PROCEEDINGS of the 1976 Technical Session on Cane Sugar Refining Research

January 23–25, 1977
New Orleans, La.

Science and Education Administration
U.S. Department of Agriculture

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FOREWORD

This technical session was sponsored jointly by the Cane Sugar Refining Research Project, Inc. and the Southern Regional Research Center, U. S. Department of Agriculture. The program was assembled by Dr. Frank G. Carpenter and Dr. Margaret A. Clarke; The Conference Coordinator was Shirley T. Saucier, and these Proceedings were edited by Dr. Margaret A. Clarke.

This is one of a series of technical sessions held every other year to provide for an exchamge of information among technical leaders in the cane sugar industry, and to report on research and recent developments of benefit to the cane sugar refining industry.

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Cane Sugar Refining
Research Project, Inc.

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THE ACTIVATION OF BONE CHARCOAL BY THERMAL AND CHEMICAL TREATMENT
PART I. THE EFFECT OF HIGH KILN TEMPERATURE ON BONE CHARCOAL STRUCTURE

By J. Colin Abram and Michael C. Bennett[1]

(Presented by Michael C. Bennett)

ABSTRACT

The structural changes which occur when char is kilned over the temperature range 400°C to 1000°C have been investigated by physical and chemical methods. The physical methods used were an electron microscope examination for structural changes combined with X-ray diffraction for crystallographic changes; gas adsorption and desorption, porosimetry and gas phase density measurements for surface area and porosity changes. The chemical methods employed were adsorption from solution techniques to measure the changes in extent of the hydroxyapatite and carbon surfaces.

The increase in decolorizing activity with increase in kilning temperature has been attributed to an increase in porosity in the range of pore diameters from 40 nm to 100 nm.

INTRODUCTION

The phenomenon of "overburning" has been recognized for well over 100 years and accurate descriptions of the behavior of overburned char date back to 1869. The characteristic effects (inferior liquor decolorization, "sulfurous" odor and high pH) were taken to indicate a chemical breakdown of char structure, and striking evidence in support of this view was provided when the Bone Char Research Project showed a catastrophic loss of B.E.T. nitrogen surface area on heating new char above 550°C (9)[2]. The matter was apparently concluded when the B.C.R.P. identified the optimum temperature for color removal as 500°C and for ash removal as 560 °C (8).

It was therefore somewhat surprising when a sample of nearly new char kilned at a temperature of about 630°C was found to be extraordinarily active in decolorization, a property which was retained on subsequent kilning. The various explanations put forward drew attention to the improvement generally found in new char over its first few cycles, and it seemed likely that the effect might be attributed to an acceleration of this improvement. Nevertheless, if the B.C.R.P. findings were applicable to this char, here was an example of a severe loss of B.E.T. nitrogen surface area accompanied by a marked gain in decolorization activity.

Meanwhile, during the course of an investigation into the sulfur constituents of char, it was found that the decolorizing activity of Thames fine

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[2] Numbers in parentheses refer to items under "References" at the end of this paper.

grist stock increased considerably on heating at 750° C, provided the char was washed either with water or sodium hydroxide solution after the high temperature treatment (7).

In bottle shaking decolorization tests, the sodium hydroxide wash was found superior, the effect apparently being related with the removal of a particular sulfur component from the char surface.

As a result of this observation, more extensive column decolorization tests were undertaken on two stock chars, each kilned at 550°C, 750°C, and 950°C, followed by water and sodium hydroxide washing. The liquor decolorization results indicated that the higher temperatures gave gains of 50% to 100% in the volume of liquor passed to a given bulk color, and that while the washing was absolutely essential for the activation, water was superior to sodium hydroxide. Since no sulfur component was affected by water washing, it seemed unlikely that the high temperature activation concerned changes in the sulfur constituents of the char.

Analysis of the water washings from stock chars kilned at temperatures ranging from 400°C to 1000°C shows that below 700°C the main constituent is calcium sulfate, above 700°C, calcium hydroxide.

There can be little doubt that the elutable calcium hydroxide is largely responsible for the characteristics of overburned char, as these effects can be simulated by adding lime to a normal char. A poor liquor decolorization performance might be attributed to color formation during the test. Sulfide ions are released from chars containing a particular "reducible sulfur" component only in liquors of high pH, so that the "sulfurous" odor does not arise when using chars washed free from lime.

It would therefore appear that the removal of lime by water washing also removes the 560°C limit on regeneration temperature, and it is interesting to compare the liquor decolorizing activity of a char kilned over the range 400°C to 900°C with and without water washing.

Results for bottle-shaking decolorization tests are shown in table 1.

The improvement in liquor decolorization activity with increase in kiln temperature for washed chars is general for all char types studied, but the magnitude of the effect varies considerably. This is shown in figure 1.

TABLE 1--Bottle shaking decolorization tests on washed and unwashed char samples

	% Decolorization					
Kiln temperature °C	400	500	600	700	800	900
Unwashed	41	44	33	32	9	-47
Washed	53	52	49	57	73	73

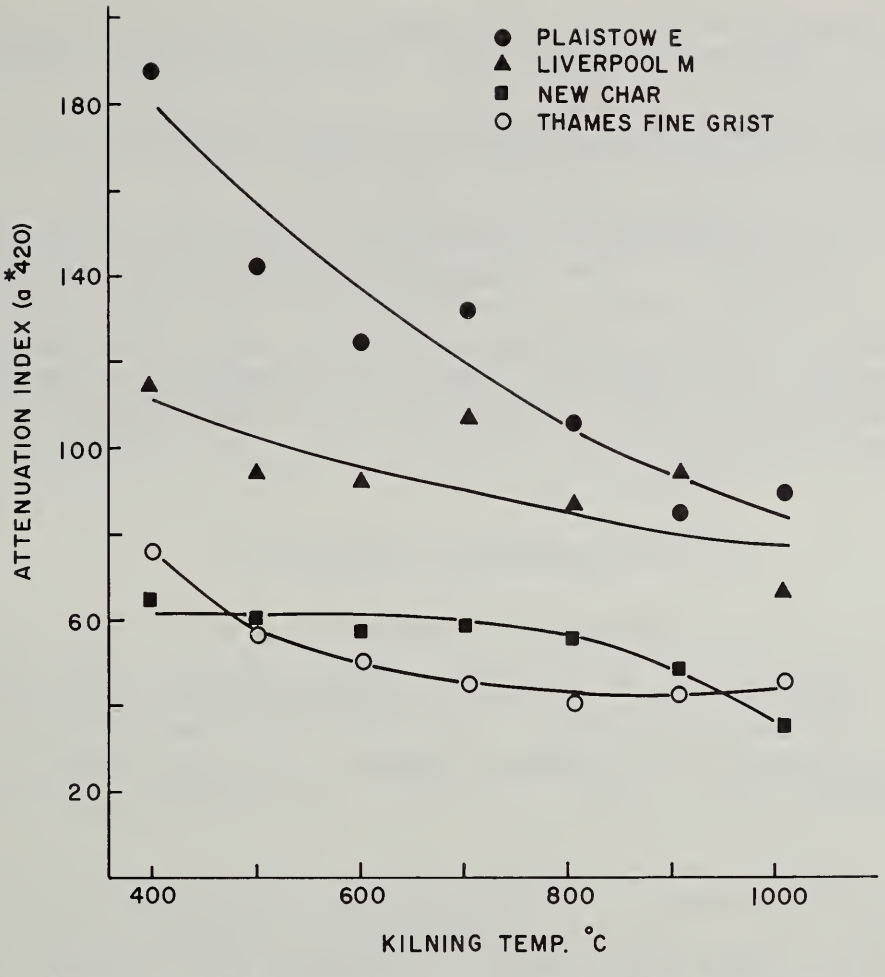

FIGURE 1--Decolorization by new chars and stock chars, kilned 400°C-1000°C and washed.

It is clear that a considerable improvement in activity can be gained by heating char at temperatures many hundred of degrees above those in current use.

This does not necessarily mean that the optimum temperature for regular revivification is also at this high level, nor has it been established that the gain in activity is achieved without loss of char life. The important point is that activation is possible: the problem to be solved concerns the nature of the physico-chemical changes which occur on heating and the identification of those which lead to activation.

This paper presents the results of investigations into structural changes which occur in char over the temperature range 400°C to 1000°C. The following techniques have been employed:

1. electron microscope examination for evidence of structural changes.

2. X-ray diffraction for evidence of crystallographic changes.

3. surface area changes by gas adsorption, and porosity changes by mercury porosimetry, nitrogen desorption and gas phase density determinations.

4. surface area changes in the hydroxyapatite component and in the carbon component of char by adsorption from solution.

The results of these investigations indicated methods by which the hydroxyapatite and carbon components of char might be independently activated or deactivated, and experiments are described where this has been achieved: the evidence suggests that these two components provide quite different adsorption sites. The high temperature activation apparently concerns an increase in porosity in pores of 40 nm to 100 nm diameter and an increase in the chemical activity of the carbon surface.

PHYSICAL METHODS OF EXAMINATION

Electron Microscopy

The technique employed after obtaining suitable samples has been published in detail (4). It involves preparing 40 nm thick microtome sections of individual char particles bedded in a Bedacryl polymer block for direct electron microscope observation.

Figures 6 to 11 show electron micrographs at 100,000x magnification of samples of new bone char heated in nitrogen at temperatures ranging from 500°C to 1000°C for 1 hour. At the lower temperatures the hydroxyapatite structural elements are similar to those in bone (4) and there is no evidence of the carbon component.

It is seen that above 700°C the thin plates disappear, to be replaced by more rounded particles which increase in diameter with increase in temperature.

This is an important finding for it indicates the possibility of an appreciable change in the porosity of the char particle, which would allow greater accessibility of colorant molecules to the internal surface. Figures 12 and 13 show electron micrographs at 100,000x magnification of Thames fine grist char heated at 550°C and 950°C: the change in structure is similar to that found in new char. Evidence of microporosity within individual hydroxyapatite platelets is indicated at arrow A in figure 12.

X-ray Diffraction

The release of large quantities of calcium hydroxide from new char heated at high temperatures suggested that the structural changes illustrated above might be a result of the chemical change from hydroxyapatite to β-tricalcium phosphate which is known to occur in precipitated hydroxyapatite around 800°C:

$$Ca_{10}(PO_4)_6(OH)_2 \xrightarrow{800°C} 3\ Ca_3(PO_4)_2 + Ca(OH)_2 \qquad (1).$$

After heating new char at 1000°C, it was possible to elute 280 μmoles of $Ca(OH)_2$ per g char, this being 35% of the maximum theoretical yield for complete conversion. The appearance of a new phase, to the extent of a 35% change, should be readily detected by the X-ray diffraction pattern of the powdered material, but no such evidence has been obtained. An X-ray diffraction examination has shown that the original hydroxyapatite pattern is retained at 1000°C with a considerable sharpening of the diffraction lines as an indication of the growth of the apatite crystallites.

In the absence of any evidence to suggest destruction of the apatite lattice, it would appear that the structural changes observed at high temperatures should be regarded as a sintering process. It is therefore to be expected that appreciable changes in surface area will occur, and the following sections describe the attempts to measure these changes.

Porosity Measurements

Samples of Thames fine grist char heated at 550°C for 1 h and at 950°C for 6 h, followed by water washing, have been analyzed for changes in porosity. A summary of the results is given here.

Table 2 shows the apparent densities of the two char samples in helium, di-n-butyl phthalate, and mercury: the bulk densities were 1.14 and 1.21 g/cm^3 respectively.

TABLE 2--Apparent densities

Sample	Density, g/cm^3 at 25°C		
	Helium	Di-n-butyl phthalate	Mercury
550°C	2.897	2.785	1.859
950°C	3.007	2.932	1.995

There is a mass loss of 5% to 10% on heating the char to 950°C and water-washing (due to carbon burn-off and removal of $CaCO_3$ as $Ca(OH)_2$), and the increase in mercury density therefore indicates a significant loss of particle volume.

Measurement of the adsorption of nitrogen, neon and argon reveals a considerable loss of surface area in the 950°C sample. The specific surface areas calculated from nitrogen adsorption data and equilibrium volumes of neon and argon adsorbed at 25 ± 0.0;°C are shown in table 3.

The Pierce interpretation of the nitrogen adsorption data assumes that the surface area is accommodated in pores of cylindrical shape. Agreement between the Pierce and BET surfaces for the 550°C sample suggests that the assumption is not unreasonable, but the large difference for the 950°C sample could be taken to indicate a marked departure from the cylindrical pore shape. Such a change is in accordance with the electron micrographs of these same char samples as shown in figures 12 and 13.

TABLE 3—Nitrogen, neon, and argon adsorption

Sample	Specific surface by sorption of nitrogen at 77°K		Apparent adsorption	
			Neon	Argon
°C	BET m^2/g	Pierce m^2/g	volume, $x10^3 cm^3/g$	volume, $x10^3 cm^3/g$
550	74.5	72.4	10.3	173
950	48.6	56.9	7.0	131

TABLE 4—Porosities

Sample	% Porosity	
	Accessible to helium	Accessible to di-n-butylphthalate
550°C	35.8	33.3
950°C	33.6	32.0

The porosities calculated from the data in table 2 are given in table 4.

It is seen that in the 550°C char, the porosity accessible to helium (0.2 nm diam) but not accessible to di-n-butylphthalate (0.7 to 0.8 nm) is 2.5, decreasing to 1.6 in the 950°C sample. Thus the overall loss of 36% of the original porosity on heating at 950°C apparently concerns changes in the micropore structure of the particle.

Further detailed examination of the distribution of porosity over a range of pore entrance diameters was made from the nitrogen desorption isotherms and from the penetration of mercury under very high pressure. The results are tabulated in table 5.

The results indicate clearly a gain in porosity for the 950°C sample in pores of entrance diameter 40 nm to 100 nm which is more than balanced by a loss of porosity in smaller pores. The size range in which the additional porosity is developed is in excellent agreement with the range measured from the electron micrographs in figure 13.

These findings may have an important bearing on the enhanced liquor decolorization activity shown by the 950°C char. The loss of nitrogen surface

TABLE 5--Distribution of porosity

Range of pore entrance diameters, nm.	% Porosity			
	From N_2 desorption		From mercury porosimetry	
	550°C	950°C	550°C	950°C
5,000 - 120	-	-	4.6	4.1
120 - 100	-	-	0.5	0.6
100 - 80	0.3	1.1	0.5	1.4
80 - 60	0.3	1.2	0.7	1.3
60 - 40	0.3	2.0	0.9	2.4
40 - 30	1.6	1.6	1.6	2.4
30 - 20	5.8	3.4	6.5	4.3
20 - 14	5.0	3.9	5.7	4.4
14 - 3	15.8	12.2	-	-

area may be quite unimportant if a large part of this is inaccessible to the colorant molecules, while the gain in macropore volume may provide increased accessibility to the useful surface.

The findings also throw some light on the relatively poor performance of granular active carbons compared with bone charcoal when B.E.T. nitrogen surface area is used as the criterion for assessing activity. Obviously a considerable proportion of the B.E.T. nitrogen surface area of activated carbons is ineffective in decolorization and inaccessible to the organic impurities found in impure sugar liquors. This can be demonstrated quite dramatically when "adsorption from solution" techniques using model adsorbate systems are employed (1).

Adsorption from Solution

Changes in the individual structural components of bone char, namely hydroxyapatite and carbon, resulting from high temperature kilning have been followed using adsorption from solution techniques.

These techniques have been described elsewhere (2, 5) and their application in evaluating char quality has been established (3, 6).

The model adsorbate molecules employed were:

1. sodium di-2-ethylhexylsulfosuccinate (OT) from water for the total surface area

2. cetyltrimethylammonium bromide (CTAB) from water for the carbon surface area

3. sodium di-2-ethylhexylsulfosuccinate (OT) from benzene for the hydroxyapatite surface area

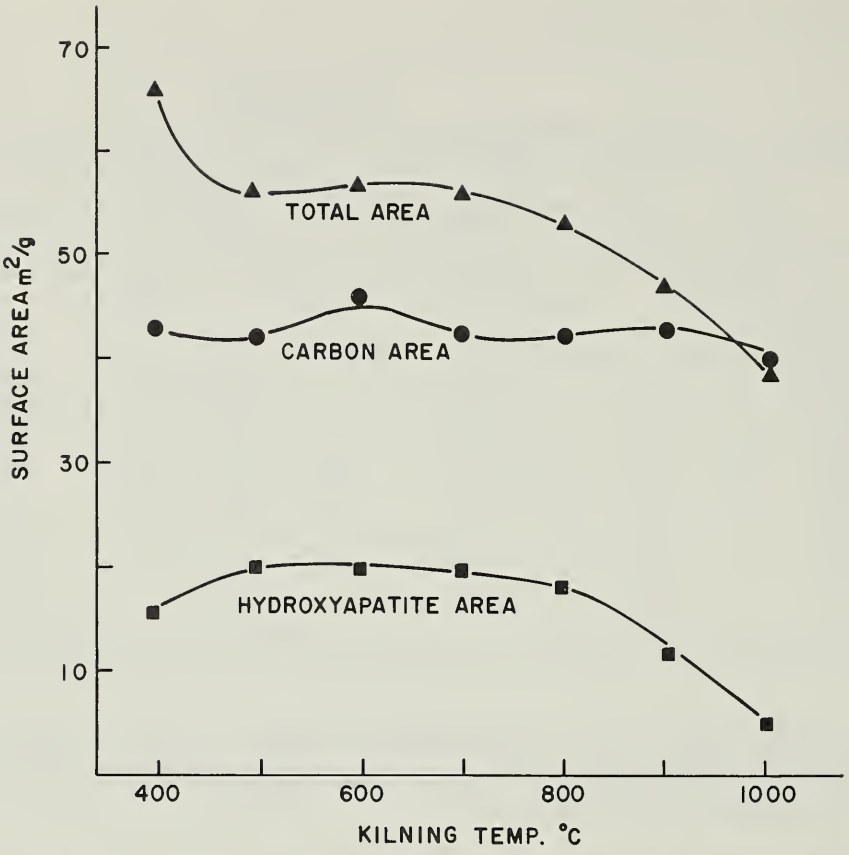

FIGURE 2—Surface areas of Plaistow E stock char.

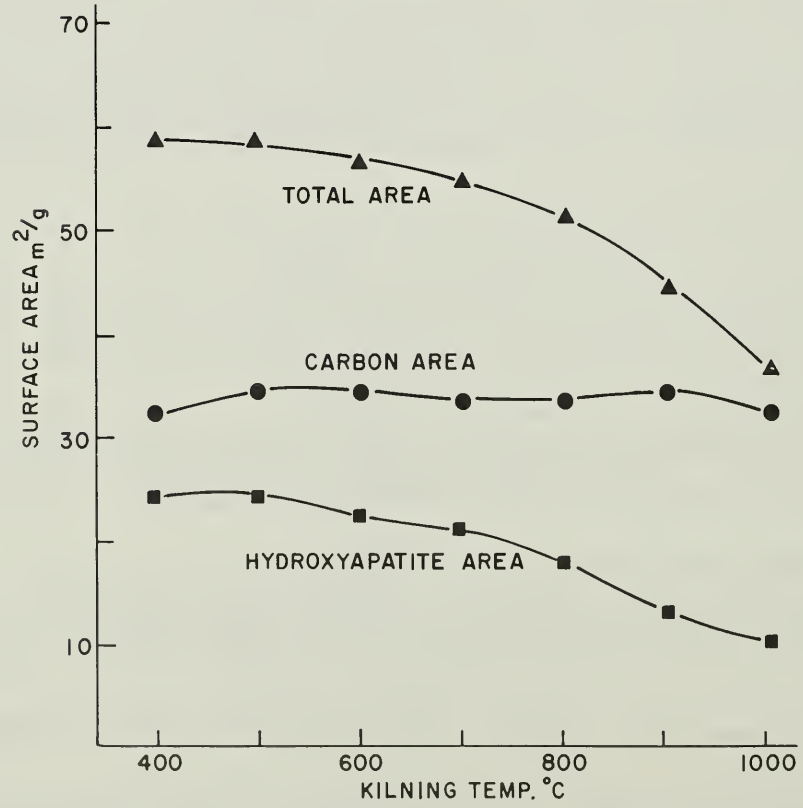

FIGURE 3—Surface areas of Thames fine grist char.

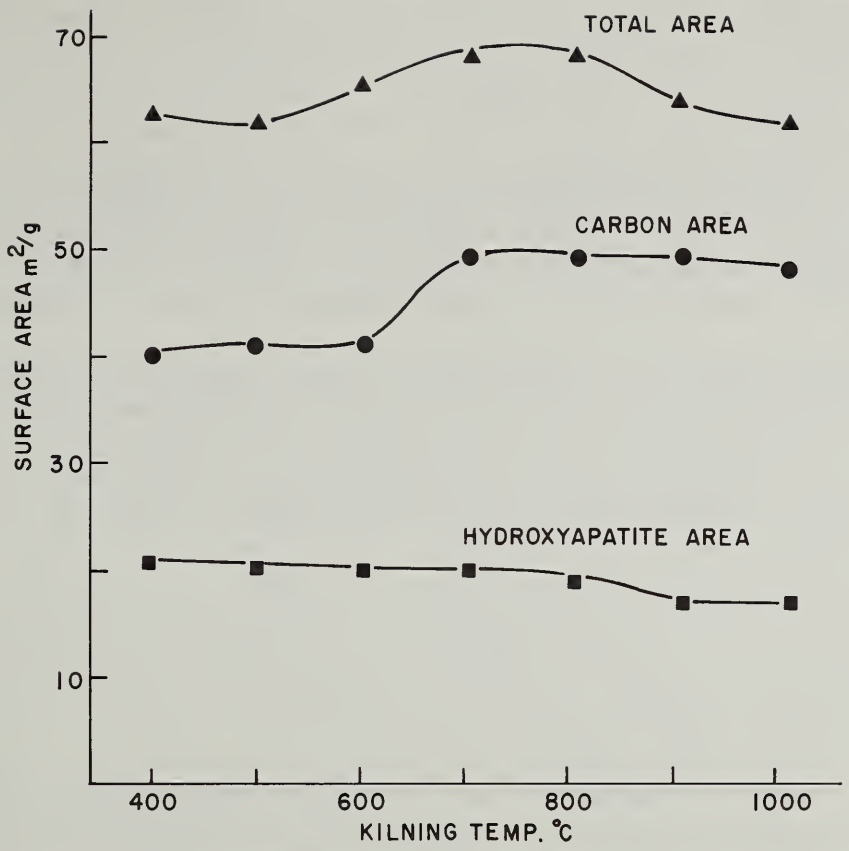

FIGURE 4—Surface areas of Liverpool M stock char.

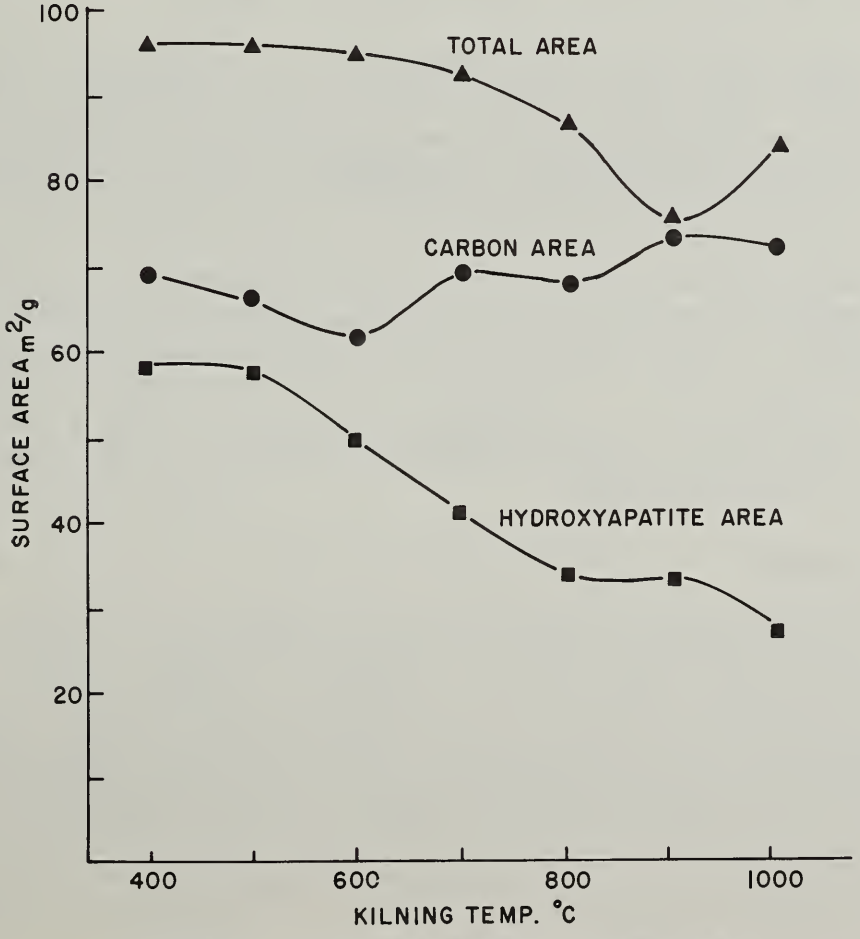

FIGURE 5--Surface areas of new char.

The results obtained on three stock chars and a sample of new char are shown in figures 2 to 5.

Total Surface Area

The stock chars kilned at 400°C have surface areas of about 60 m^2/g compared with 95 m^2/g for new char. These values are somewhat lower than the expected B.E.T. nitrogen values and the difference may indicate that part of the nitrogen surface is inaccessible to the relatively large 'OT' molecules.

All chars except Liverpool M stock show a marked decrease in surface area with increase in temperature, but the change is gradual, as suggested by the electron micrographs in figures 6 to 11, and there is no evidence of a "catastrophic" collapse. With the Liverpool char (figure 4), there is an apparent increase in surface area between 500 and 800°C and the decrease in surface area is only detected at 900°C.

Some light has been thrown on the problems posed by the behavior of new char reported by the B.C.R.P. and the behavior of Liverpool char described here: it appears that the carbon content of char is effective in delaying the structural changes until higher temperatures are reached.

Thus a sample of new char decarbonized in oxygen at 700°C was found to have a surface area of only 45 m^2/g. A sample of bone, freed from organic matter by refluxing in alkaline glycerol, and subsequently washed with water, had a surface area of 93 m^2/g, but on heating to 900°C the area decreased to 57 m^2/g compared with 75 m^2/g for new char heated at 900°C.

Since at any temperature the carbon content of the Liverpool M stock is approximately 3% greater than that of the other chars, it is suggested that the carbon constituent prevents contact between apatite units and thereby delays the sintering at elevated temperatures. Preliminary electron micrographs of the Liverpool char are in support of this view.

It is evident that the increase in decolorization activity produced by kilning at elevated temperatures is not associated with any increase in the total surface accessible to 'OT' molecules (0.57 nm^2 in cross section), but it is possible that there has been an increase in surface accessible to the larger colorant molecules. Adsorption from solution techniques, using solute molecule s larger than 'OT', might yield further information on this matter, but suitable molecules are not yet known.

The Hydroxyapatite Surface

The change in hydroxyapatite surface area with increase in kiln temperature generally follows closely the observed changes in total surface area: Plaistow E, Thames fine grist and new char all show a marked decrease in hydroxyapatite surface while Liverpool M char shows little change. This is in accordance with the view, first put forward by the B.C.R.P., that it is the structure and state of the hydroxyapatite component which determines the surface area of bone char.

The results are important for they suggest that the gains in liquor decolorization activity are not associated with changes in either the total area or the hydroxyapatite surface: attention has already been drawn to the change in pore size distribution which would lead to an increase in accessibility of larger colorant molecules. There remains, however, the possibility that the increase in decolorizing activity might be attributed to the change in nature of the carbon surface.

The Carbon Surface

There is no loss of carbon surface area over the temperature range studied with any of the chars, in spite of the considerable carbon burn-off. As in previous decarbonization experiments there is no evidence to suggest pore unblocking: increases in carbon surface area shown, for example, in Liverpool char (figure 4) at 600°C to 800°C were not found in the unwashed char samples and may be an artifact. In this connection, it is possible that the increased adsorption of CTAB, from which the increase in surface area was calculated, arises through a change in the nature, rather than the extent, of the carbon surface.

In general the results indicate a steady increase in the carbon surface area per unit mass of carbon, and therefore suggest that fresh carbon surface is exposed at the higher temperatures. The effect is perhaps similar to that which occurs in the regeneration of granular carbon adsorbents where a substantial burn-off at 900°C to 1000°C appears to be essential for proper activation.

CONCLUSIONS

The decolorization activity of bone char improves with increasing kiln temperature, provided the char is washed free from calcium hydroxide. The lime formed at temperatures above 700°C does not appear to originate in the chemical change from hydroxyapatite to -tricalcium phosphate, but perhaps concerns the expulsion of carbon dioxide from a carbonate-apatite structure. In a stock char, the lime might originate in the breakdown of carbonatation calcium carbonate present in the char, or in the expulsion of calcium ions accumulated in previous liquor cycles.

Attempts to measure changes in specific properties of char with increasing kiln temperature have shown:

1. a marked change in structure above 700°C
2. a decrease in total surface area
3. a decrease in apatite surface
4. a decrease in carbon content, but no change in the carbon surface area

The findings confirm the view that the structure of a char particle is determined primarily by the apatite component. The structural change at elevated temperature is regarded as a sintering of apatite crystallites, but there

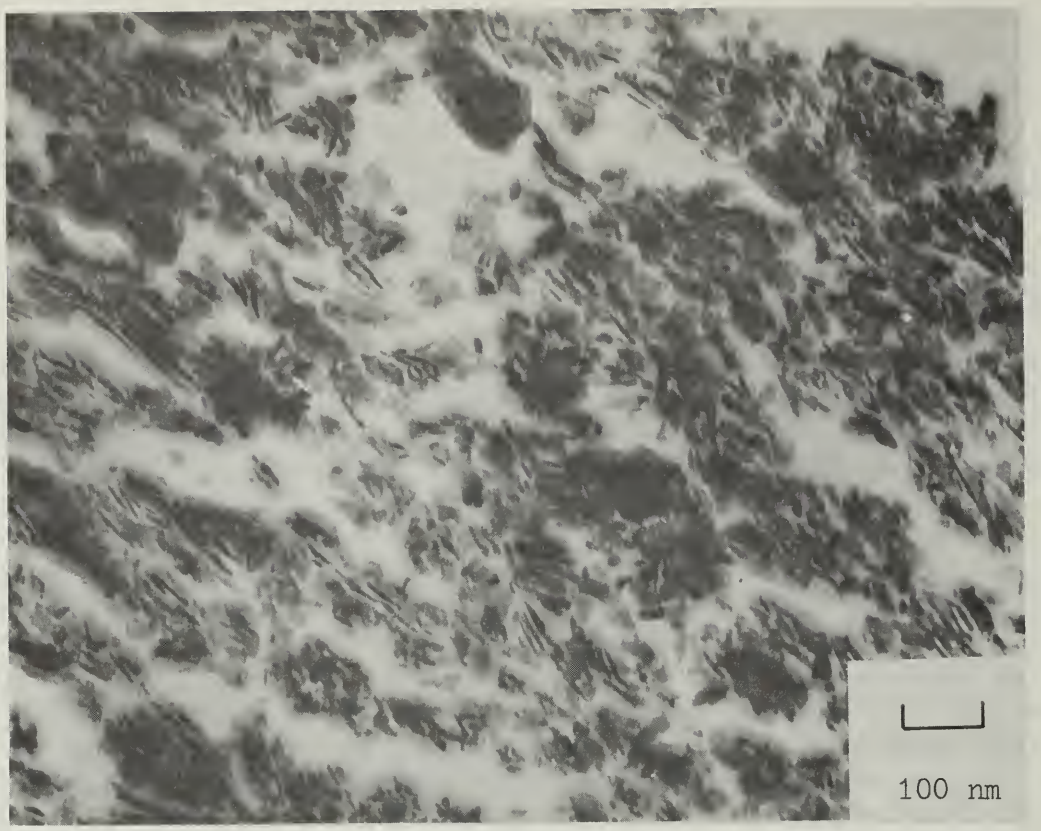

FIGURE 6--New bone char heated to 500°C.

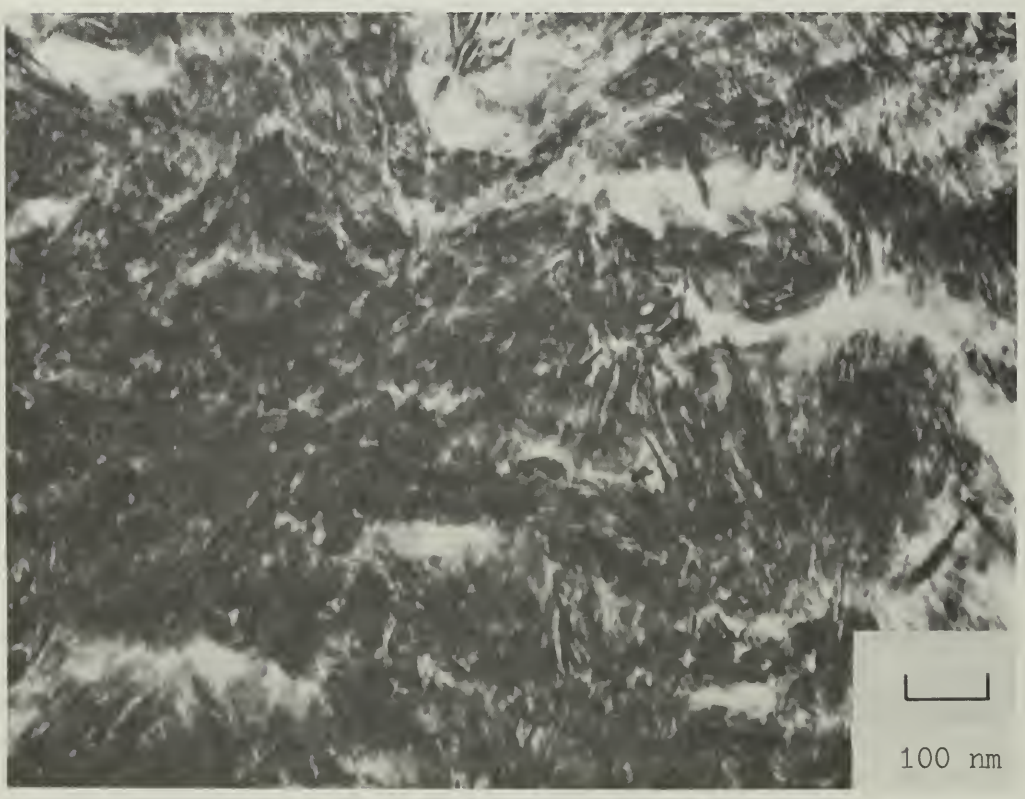

FIGURE 7--New bone char heated to 600°C.

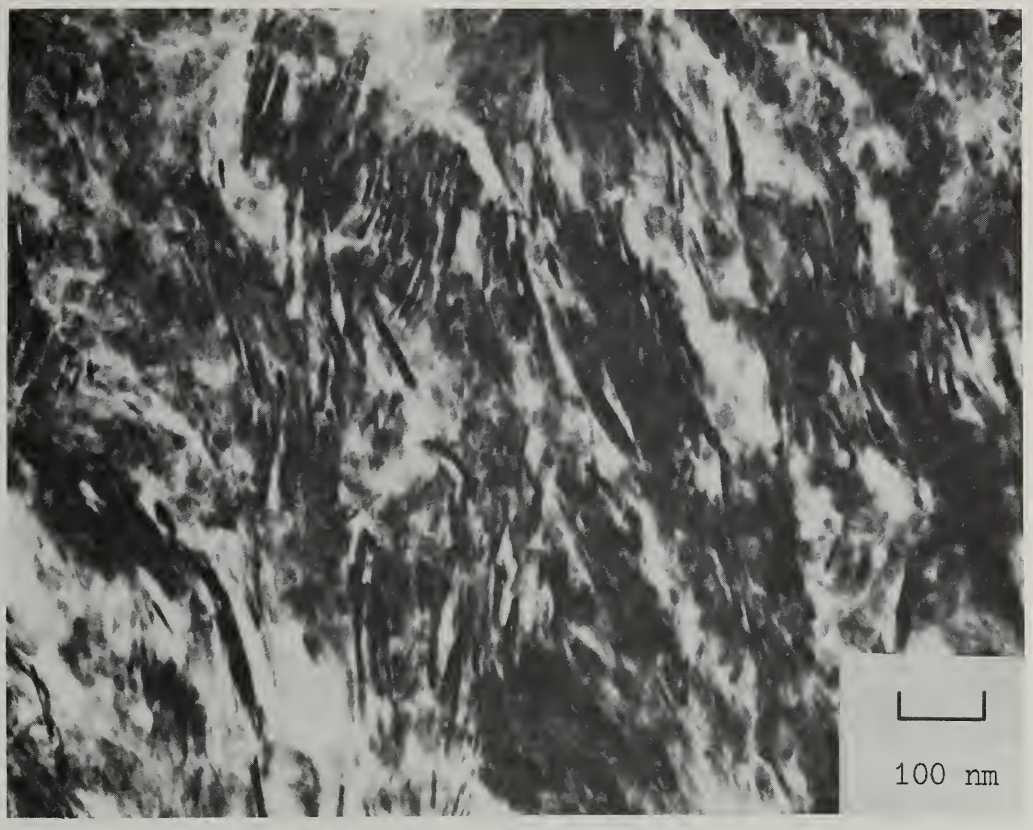

FIGURE 8—New bone char heated to 700°C.

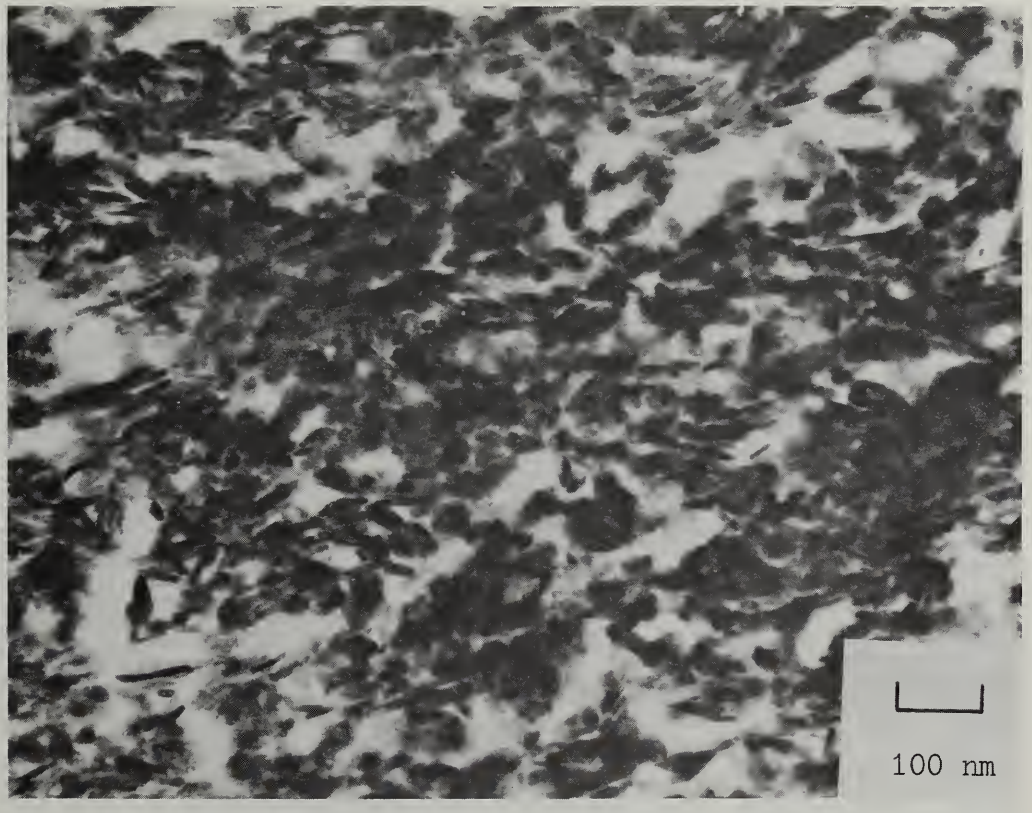

FIGURE 9--New bone char heated to 800°C.

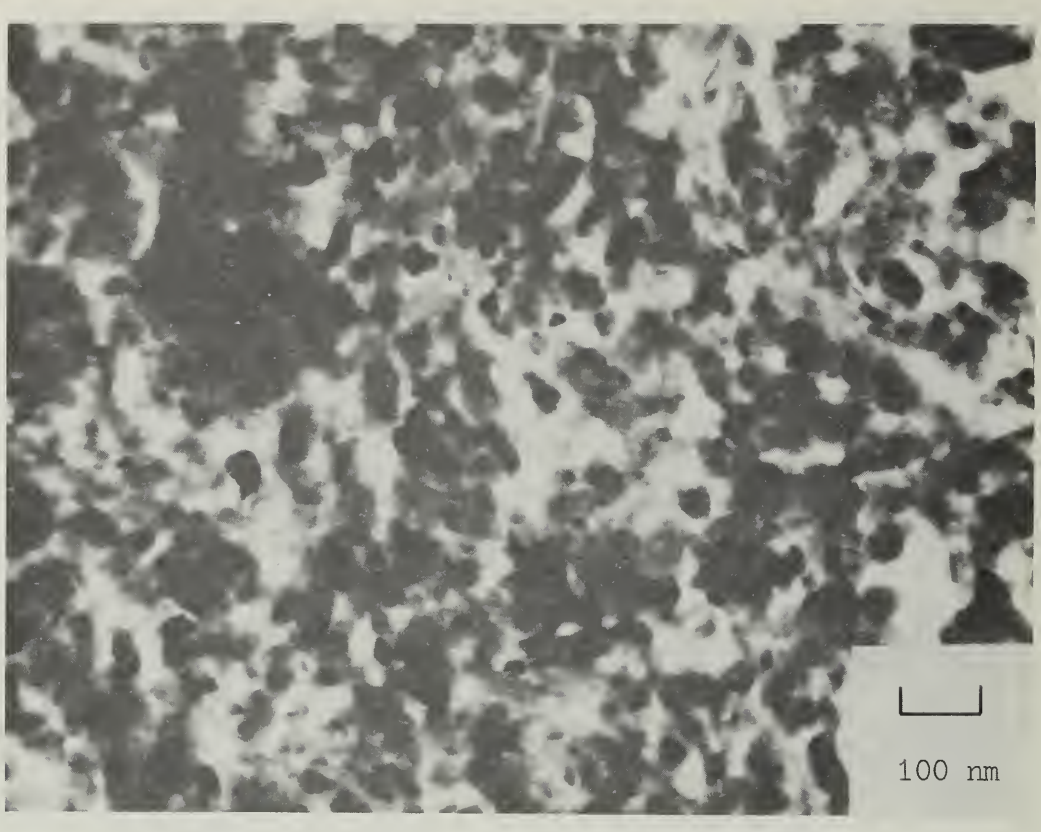

FIGURE 10--New bone char heated to 900°C.

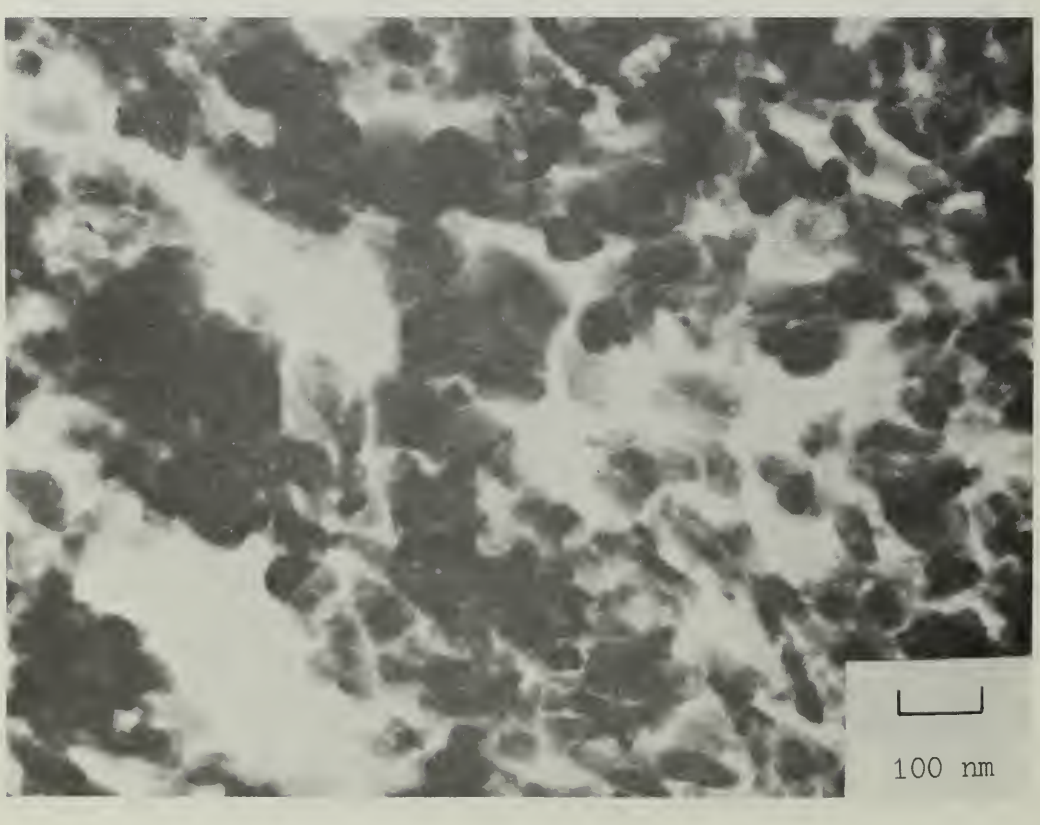

FIGURE 11--New bone char heated to 1000°C.

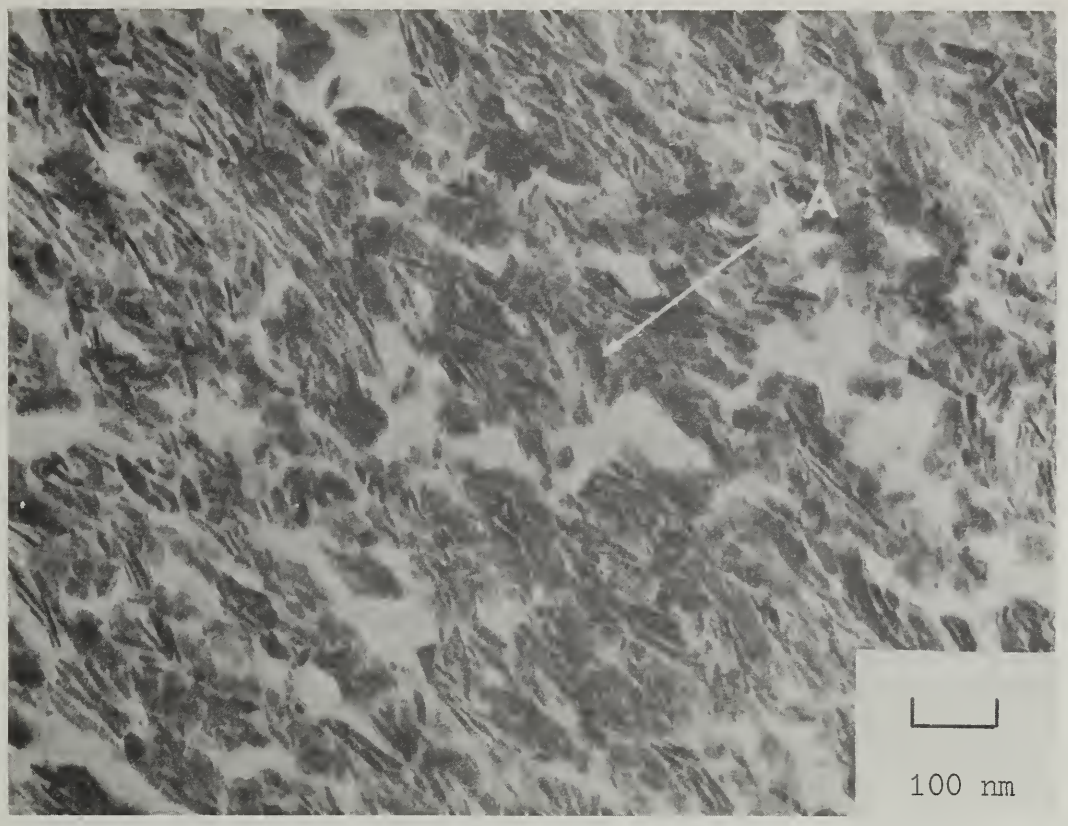

FIGURE 12--Thames fine grist char heated to 550°C.

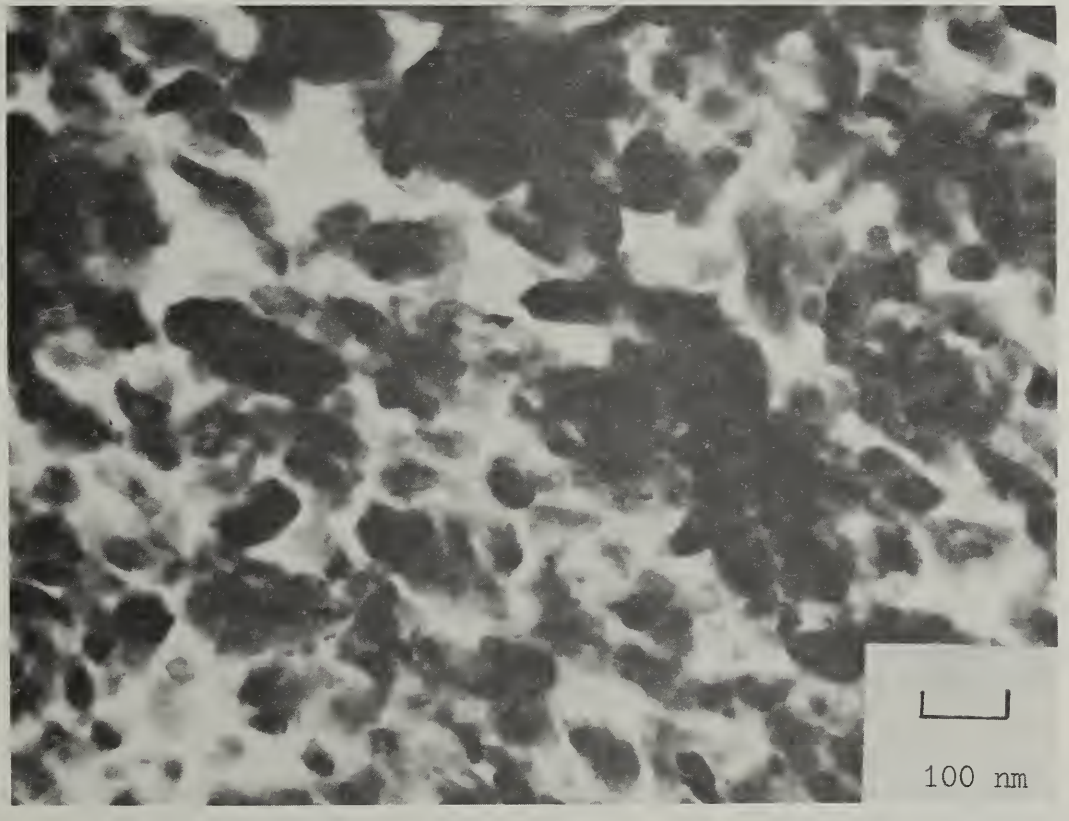

FIGURE 13--Thames fine grist char heated to 950°C.

is evidence to suggest that it is highly sensitive to the carbon content of the char: in char of high carbon content the sintering is delayed until temperatures of 900°C or above, while in decarbonized char sintering sets in around 600°C.

Although there is an apparent loss of surface area when determined by nitrogen (0.16 nm^2 crosss sectional area) and OT (0.57 nm^2, there is a gain in porosity in pores with entrance diameters in the range 40 nm to 100 nm. In fact both nitrogen desorption and mercury porosimetry techniques show an increase in total porosity in pores with entrance diameters greater than 20 nm for a stock char kilned at 950°C.

The loss of surface area resulting from high temperature kilning of char appears to occur exclusively in micropores which probably play little or no part in the decolorization process.

The increased accessibility to colorant molecules and indeed to the liquor itself might account for much of the observed activation, the effect being similar to that of a reduction in grist size.

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F. G. Carpenter (CSRRP): When bone char is first manufactured it is initially heated to a temperature that is higher than that at which it is usually regenerated. What is the effect of this initial heating temperature on the regeneration at higher than usual temperatures?

M. C. Bennett (Tate & Lyle): In the manufacture of bone char, when raw bone grist goes into the retorts it contains much organic matter. We believe this organic matter protects the hydroxyapatite crystallites against any form of sintering. However, we know that if you leave new bone for long at high temperatures, it will indeed sinter slowly. Completely degreased bone, i.e. raw bone free from all organic matter, sinters at very low temperatures.

F. G. Carpenter. You were mystified somewhat by the appearance of calcium hydroxide and the sharpening of the hydroxyapatite peaks. There is a reaction where hydroxyapatite could conceivably go to octacalcium phosphate, which would leave a calcium hydroxide left over. Octacalcium phosphate has an X-ray diffraction pattern very similar to hydroxyapatite - only one of the minor peaks is different. In the past, what was often thought to be hydroxyapatite was really a mixture of hydroxyapatite and octacalcium phosphate. They are of so nearly the same crystal structure that they can actually form interlayered crystals: one layer of hydroxyapatite and one layer of octacalcium phosphate. At usual temperatures, it is considered that octacalcium phosphate is not as stable as hydroxyapatite and the reaction should therefore go towards hydroxyapatite, but at high temperatures, and under the conditions of many absorbed components from the sugar, the equilibrium may be pushed so that octacalcium phosphate could be formed. This would result in a sharpening of the peaks and the production of calcium hydroxide and would explain the effect you observe.

M. C. Bennett: Thank you, Frank. I think that we should go back and review the basic information and see if that could be an explanation for some of the observations we have made.

G. W. Muller (SIT): I wish that the late Dr. E. P. Barrett could have heard this paper in which many of his ideas are proven. He believed in high porosity and built it into his Synthad.

In the 30's, National Sugar Refining Co., in making liquid sugar, burned char at very high temperatures. They had three chars, and used their best grade burned at over 1150°F in old cast iron kilns. It was, I should say, 150°F to 250°F higher than the normal char burning temperature. The chars were of extremely high alkalinity - we were concerned about that. We made possibly the finest liquid sugar in the New York market at that time by washing that char after burning, sweetening it on, and producing a top-quality commercial liquid sugar, from what was then over-burned char. We also made the finest soft sugar that we ever produced, primarily from clarified and char-filtered affination syrup. That material went over the over-burned char over which any normal liquor would certainly have gotten a greenish color.

K. R. Hanson: In view of recent developments in the long range supply of bone char and granular carbon, would you like to comment on whether this represents research on an endangered species?

M. C. Bennett: Speaking from the point of view of British Charcoals & Macdonalds: in our opinion bone char is not going to disappear for many, many years to come. In recent years our sales have increased appreciably and, in fact, today our business volume is better than ever before in our history. We do not see any end to the use of bone char and for this reason we are expanding and modernizing our facilities in Greenock. We understand and believe we have solved the problem regarding the supply of bones. We recognize the pressure from competitive adsorbents and we understand the competition within our own company from competitive systems. Nevertheless, we feel that bone char is a major sugar refining aid, and that it is likely to remain that way for a long time.

N. H. Smith (C&H): Can you add to char carbon that will have the same activity as the original carbon?

M. C. Bennett: You can't just add carbon. You have to put some organic material in the char and reduce it back to carbon. The answer is, yes you can do it, but only indirectly. We have used hydrocarbons, e.g. hexane, successfully for this purpose.

C. C. Chou: What would be the effect of the high temperature regeneration on the ash removal and buffering capacity of the char?

M. C. Bennett: The regeneration procedure has never been used in commercial practice, and I don't believe we have much data on the ash removal.

C. C. Chou: As you remember, bone char does three things: it removes color, ash, and it has buffering capacity. These aspects have to be considered too. What was the objective of this study?

M. C. Bennett: We were seeking the truth! In all seriousness, this was only a part of a very large research project to investigate how we could activate bone char beyond all expected performance criteria. This paper is only part of our research report, and it refers only to the purely structural changes. The chemical changes are described in another part of this work which I trust will be presented in due course. The fact of the matter is that we continually encountered results that could not be explained by the published literature, so we decided to have a closer look at it ourselves just to see if there could be any possible value to us as sugar refiners. I believe the answer is that it did not turn out to be of commercial value to us as sugar refiners, but perhaps we should have tried a little harder. However, I think that our efforts at least allowed us to explain the hitherto unexplained.

FRACTIONATION OF SUGAR COLORANTS BY HIGH PRESSURE LIQUID CHROMATOGRAPHY

By Norman H. Smith

ABSTRACT

A high pressure liquid chromatographic technique has been applied to fractionate sugar colorants. In contrast to gelpermeation methods which yield few color peaks, HPLC separates over a dozen colorants.

Applications of the new technique are presented to illustrate its usefulness. These include variations in colorant composition among individual strikes of raw sugar, the role of enzymatic browning in sugar colorant composition, and the colorant composition of granulated sugar.

INTRODUCTION

Many laboratories have applied gel filtration to fractionate sugar colorants. Most often a chemically modified dextran, such as Sephadex G-10, has been used for this purpose. Typically, three fractions are obtained[2]. The first contains high molecular weight colorants completely excluded by the gel. In addition to colorants, the second fraction also contains the sugars present in the sample. The third fraction contains highly pH-sensitive colorants which are slowly eluted, at least in part, because of adsorption by the Sephadex. The purpose of this paper is to present a fractionation procedure based on differences in polarity, rather than molecular size, and examples which illustrate its application in various phases of colorant research.

APPARATUS AND METHODS

Recently our laboratory purchased a high pressure liquid chromatograph (HPLC) for use in sugar analysis. In this system, a pump provides flow rates up to 10 ml/min at pressures up to 6000 psi. Solids in the eluate are monitored with a differential refractometer.

By replacing the refractometer with a spectrophotometer and using a column containing an appropriate packing material, a system suitable for fractionating colorants was developed. A flow diagram is shown in figure 1.

A spectrophotometer equipped with a flow cell of 0.3 ml volume was available from apparatus previously used in gel filtration studies[3]. A wide range of sensitivities was available, with satisfactory results at a full scale absorbance as low as 0.01. The desired resolution of colorants was obtained

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[1]Senior research chemist, California and Hawaiian Sugar Co., Crockett CA 94525.

[2]Kennedy, A. M., and Smith, P. 1976. Colour in refineries. Proc. Sugar Ind. Technol. 35: 156-160.

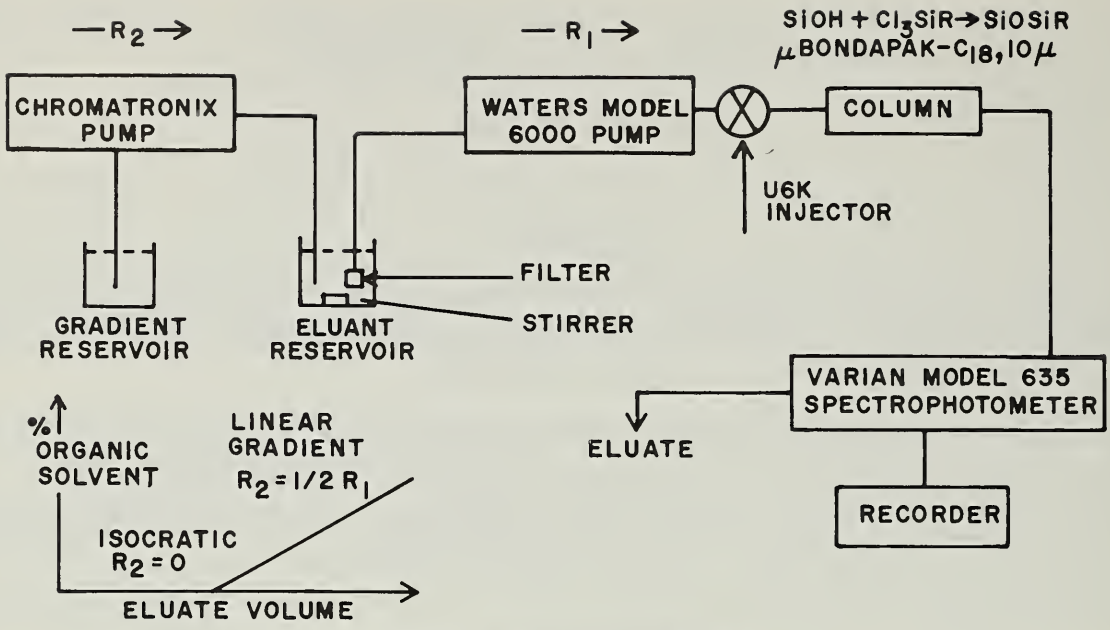

FIGURE 1--Flow diagram.

using an acidic solvent system. Because many sugar colorants are highly pH-sensitive, their detection in the acidic eluate requires measurement in the ultraviolet region of the spectrum. This region provides increased sensitivity for all of the colorants, compared to measurements at 420 nm. Thus, the presence of substances is monitored, rather than the contribution of each colorant to visual color at pH 7.

The column used throughout this study, 1/4" i.d. x 30 cm long, contained μBondapak-C_{18}, the Waters Associates trade name for a non-polar packing material. The packing is made by reacting an organosilane with silica of 10μ particle size, to provide a monomolecular layer of octadecyltrichlorosilane. The chemical bond is compatible with organic and aqueous solutions from pH 2 to pH 9. The high efficiency of the column is due to the small average particle size and narrow particle size distribution. Because of a "balanced density" packing method employed by Waters Associates, this material is supplied only as packed columns.

Because of the small particle size of the column packing, eluants and samples required filtration prior to fractionation. A membrane filter of 0.45μ pore size was used for this purpose. In initial runs, especially where gradients were employed, there was excessive recorder noise due to gas bubble formation in the eluate. This diffuclty was overcome by vacuum degassing the warmed eluants prior to filtration.

Reverse Phase Gradient Elution

A reverse phase system was employed, following the supplier's recommendation. Passing a polar solvent through the low polarity packing elutes the most

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[3]Smith, N. H. 1975. Gel filtration for determining efficiency of color removal in processed raws. Proc. Tech. Sess. Cane Sugar Refinin. Res. 1972: 1-22.

polar substances first. Elution is retarded by increasing the polarity of the solvent, as outlined in table 1.

TABLE 1--Effect of changes in solvent polarity

Normal vs. reverse phase		
	Normal phase	Reverse phase
Packing polarity	High	Low
Solvent polarity	Low to Medium	Medium to High
Sample elution order	Least Polar First	Most Polar First
Effect of increasing solvent polarity	Reduces Elution Time	Increases Elution Time
Effect of gradient		
Initial solvent: dilute acetic acid (highly polar)		Prolonged elution time for 1st colorant (most polar)
Increasing concentration of gradient solvent (decreasing solvent polarity)		Decreasing elution time for less polar colorants

The need to apply gradient elution will be discussed later. The effect of the gradient is stated in table 1. Gradients, that is reproducible transitions in solvent composition, were achieved simply by pumping a second solvent into the eluant reservoir during the fractionation. The shape of the gradient curve is determined by the initial volume and the relative rate of solvent transfer into and out of the eluant reservoir. Linear gradients used in this study were obtained by using an eluant rate twice the gradient solvent rate. Generally the gradient was preceded by a period of isocratic operation (constant solvent composition).

The main advantage of the HPLC system is in the large number of colorants that can be separated. A disadvantage lies in the susceptibility of the column to failure. Both solvent and sample must be filtered through a 0.45μ filter to avoid column pressure buildup. Physical shock, such as that caused by sudden pressure increases when the system is turned on, can cause compaction of the packing, also leading to excess pressure buildup. The column used in this study eventually plugged completely, prematurely terminating the series of tests in progress. The following examples illustrate the progress made prior to that time.

SAMPLES

Much of our recent work has beeen directed toward the colorants which survive the refining process or are produced during processing and thus contribute to refined sugar color. Since the colorants in granulated sugar are too dilute for detection in a fractionation, an attempt was made to prepare a sample of concentrated colorants representative of those in granulated sugar. To this end, a large quantity of granulated sugar solution was decolorized by passage through a bed of the non-ionic resin Amberlite XAD-2 under acidic conditions, which favor color removal. After the resin was washed to reduce the sugar content, the adsorbed colorants were recovered by extraction with methanol. The methanol solution was then adjusted to neutrality and concentrated to a small volume. The procedure was repeated on this concentrated colorant solution to remove residual sugar, yielding a sample with a^*_{420} (pH 7) of nearly 900.

Samples of "A" strike raw sugar from various Hawaiian factories were obtained for studies attempting to correlate ease of color removal in refining with the presence of particular colorants. Other samples were drawn from refinery process streams so that changes in colorant composition during refining could be followed. Finally, there were samples of cane juice produced under normal conditions, and where enzymatic browning was inhibited by heating the cane prior to extraction.

A detailed evaluation of the data obtained in the above studies is beyond the scope of this presentation. The purpose here is to illustrate the fractionation system with relevant examples. Nevertheless, some observations can be made which emphasize the usefulness of the techique.

HPLC OF SAMPLES

A series of isocratic fractionations of concentrated granulated sugar colorant (GSC) are shown in figures 2 and 3. The eluant was 2% aqueous acetic acid containing a varied percentage of methanol. Peaks are identified numerically to indicate the shift to slower elution as the percentage of methanol is decreased.

At high methanol percentages, all of the colorants are eluted within a reasonable time, but those of short retention time are unresolved. When lower methanol concentrations are used, peaks of intermediate retention show improved resolution. At very low alcohol concentrations, the first colorants eluted are adequately resolved, but later ones are eluted impractically slowly and broadly.

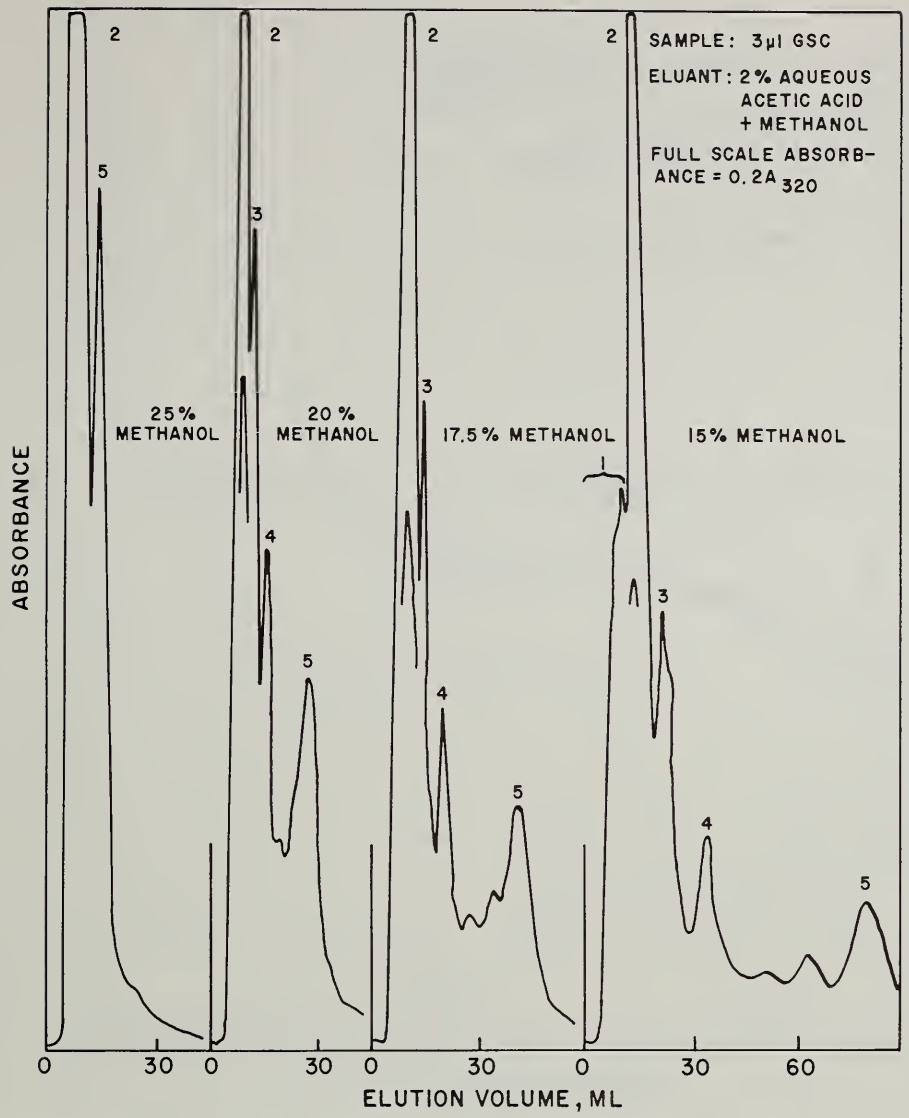

FIGURE 2--Isocratic fractionation of granulated sugar content.

There appears to be no one eluant composition which provides suitable separations over the whole range in a reasonable time. By resorting to gradient elutions more adequate separations can be achieved at the expense of simplicity and reproducibility.

In figure 4 a methanol gradient fractionation of the GSC is compared with that of granulated sugar. Inclusion of the latter illustrates the difficulty of fractionating very light colored samples, even when monitoring in the ultraviolet region of the spectrum. The presence of peaks is suggested but their magnitude approaches the overall noise level shown on the recorder.

Gradient elution permits a resolution of early peaks similar to that obtained at similarly low methanol concentrations under isocratic conditions. The right end of the GSC curve is produced at a relatively high alcohol concentration, required to ensure complete colorant elution.

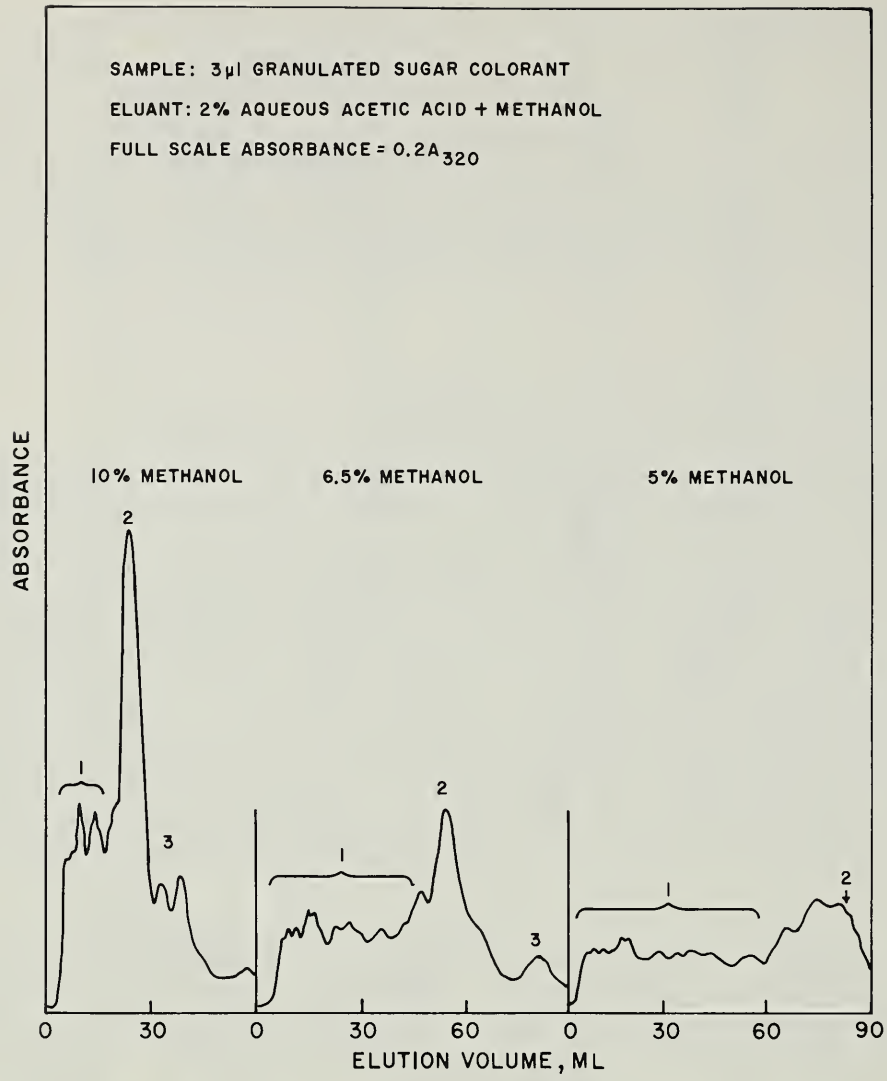

FIGURE 3—Isocratic fractionation of granulated sugar content.

A comparison of the two curves suggests that the principal peak of GSC is indeed present in the original granulated sugar. The comparison also illustrates the effect of sugar itself. Because of refractive index changes which affect the collimation of the light beam, the eluted sugars produce a response which may mask the presence of color peaks if they are present. In this instance, since the GSC does not produce peaks in the sugar response region, we may assume that those in the curve for granulated sugar are due to refractive index effects only.

HPLC of Sephadex Fractions

A portion of granulated sugar colorant was fractionated on Sephadex G-25. Eight gel fractions (I to VIII) obtained with neutral elution were then subjected to isocratic HPLC fractionation. The eluant was 33% formamide containing 0.5% acetic acid. In addition, another portion of GSC was subjected directly to HPLC under the same conditions. The resulting fractionation pattern is shown in Figure 5. Peaks are lettered for comparison with those seen

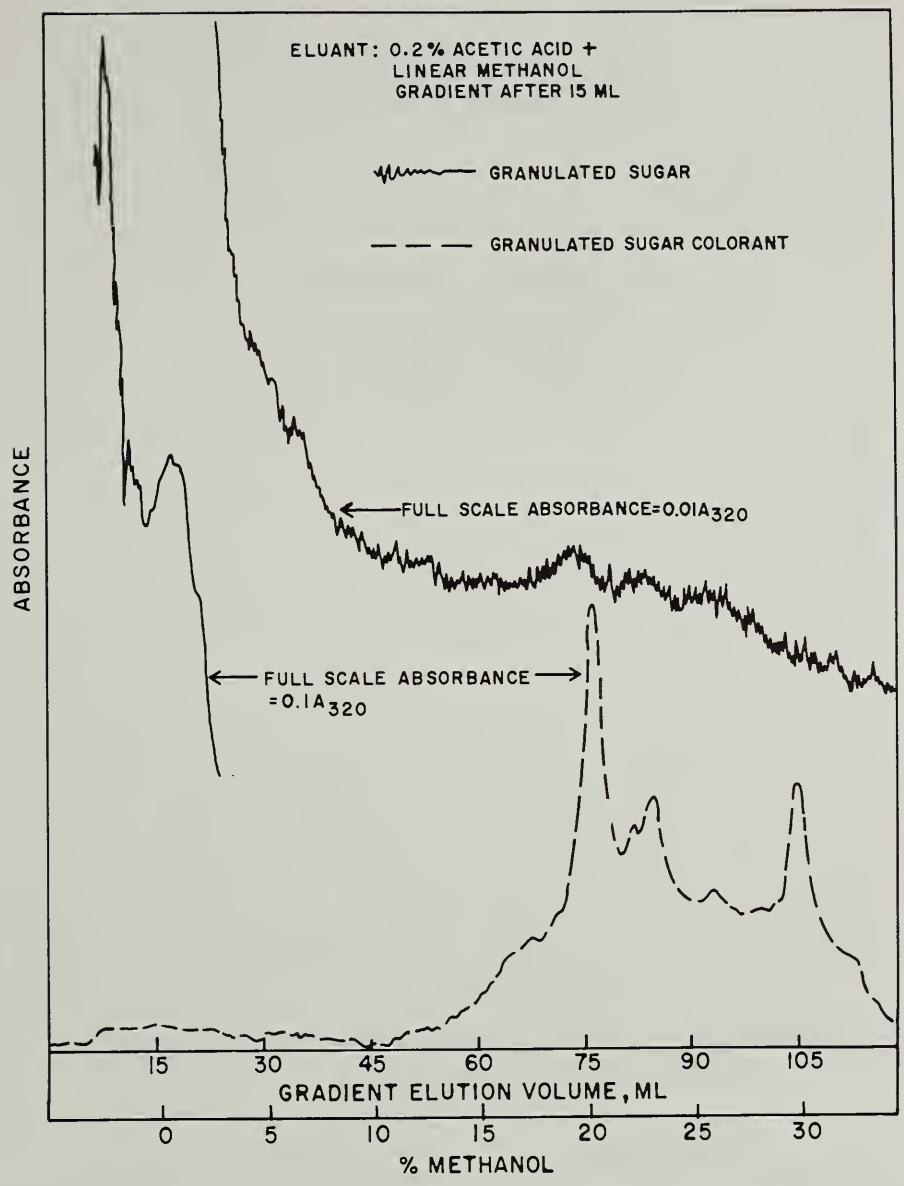

FIGURE 4--Gradient elution of sugar colorant.

in the HPLC fractionation of the Sephadex eluates, the first three of which are included in figure 5.

The A peak is not resolved in the GSC curve, but is the only constituent found in the first three Sephadex fractions.

Curves for the remaining Sephadex eluates are shown in figures 6 to 9. Changes in colorant composition are evident as the gel filtration proceeds. Except for the first Sephadex peak, (A), representing high-molecular weight colorants, it is apparent that individual colorant fractions produced by gel filtration on Sephadex are complex mixtures. Each colorant appears in a series of contiguous Sephadex fractions; i.e., peak I is seen in fractions VI, VII, and mostly in VIII. Peak C was not resolved in the GSC curve but is seen as one of the major peaks of fraction VII and in fraction VIII. It should be noted that the retention times for peaks H to L for the Sephadex fractions varied and were less than in the GSC fractionation, possibly because of gradual changes in column conditions.

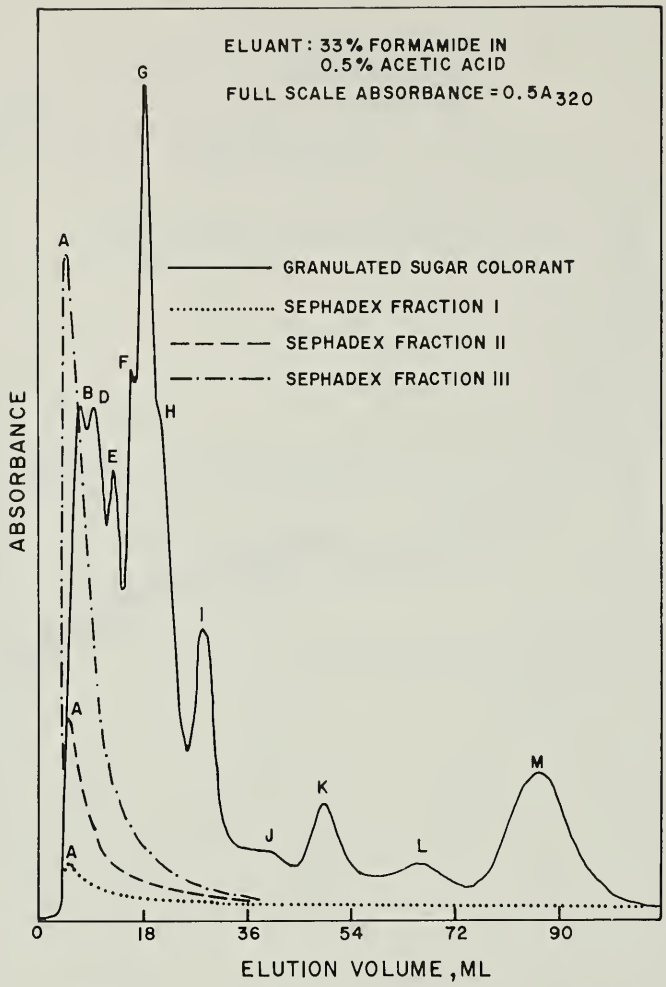

FIGURE 5—Fractionation of granulated sugar colorant and Sephadex fractions.

Comparison of Process Streams

In a previous example where the methanol concentration was varied in a series of isocratic fractionations of GSC, it was seen that at low methanol concentrations, high resolution of early peaks was obtained. Isocratic operation at 6.5% methanol and 2% acetic acid was selected to follow the content of the major peak of GSC through the refining process. A series of samples representing whole raw sugar, and liquor just prior to and after char treatment, are compared with GSC. The results, shown in figure 10, also compare GSC alone and added to refined sugar. The latter comparison again illustrates the effect of sugar solids. It also demonstrates that colorant retention is essentially independent of total solids concentration.

The buildup of the major GSC peak, (peak 2 of figures 2-4 at 51 ml elution volume) during processing is apparent. Peaks at 42 ml and 78 ml do not show a corresponding buildup. It is of special significance that the main colorants found in GSC are also found in raw sugars.

Another application of colorant fractionation techniques is based on a comparison of raws from different sources. Samples of individual "A" strike

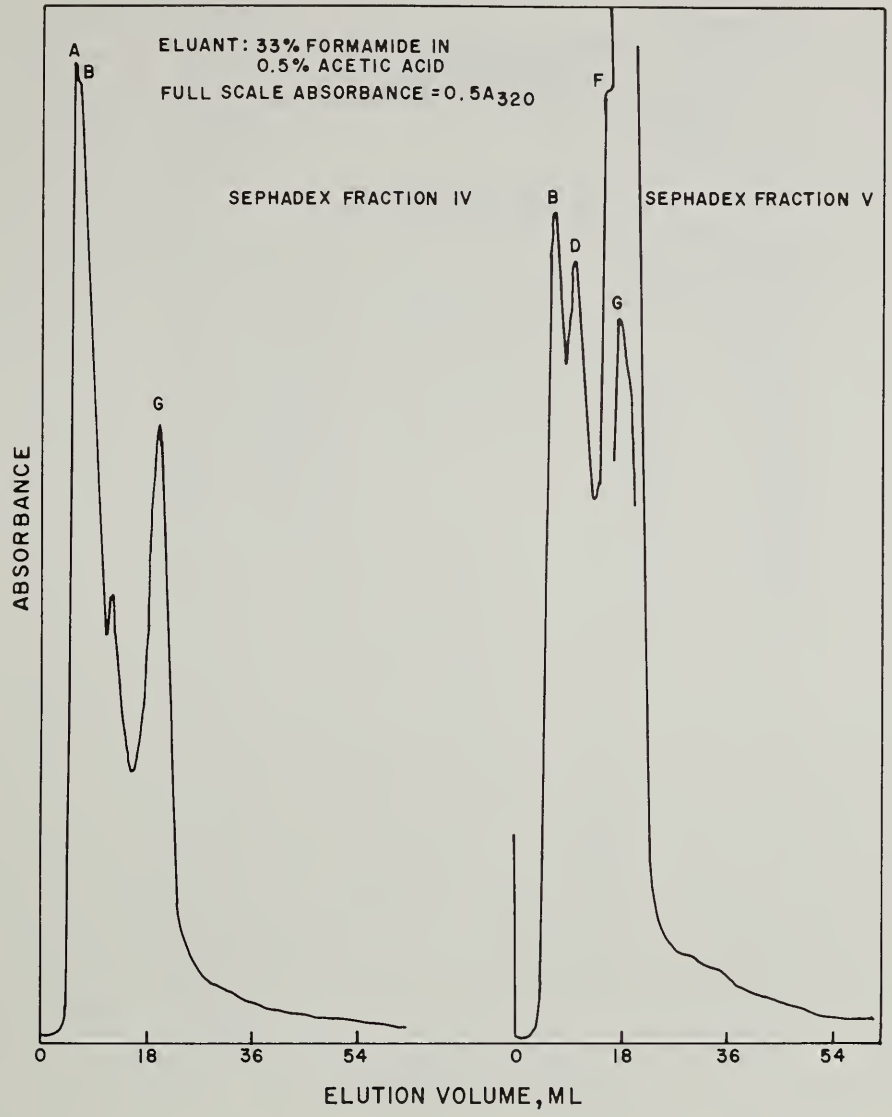

FIGURE 6--Fractionation of Sephadex fractions.

raws from different Hawaiian factories were fractionated using a methanol gradient. The results are shown in figure 11.

About a dozen well defined colorant peaks can be seen. Duplicate runs on the same sample show good reproducibility. The differences appear to be quantitative rather than qualitative. Some colorants show a fairly constant contribution to sugar color (peaks at 70 ml) while others (peaks at 50 ml) appear to be contributors to the darker color of some raws.

Enzymatic Browning Studies

The effect of heating cane prior to grinding on juice color is offered as a final area of study to illustrate the HPLC technique. In 1975, tests were made using the "mini-factory" facilities at the HSPA Laboratory in Honolulu. At that time fractionation on Sephadex G-25 indicated that enzymatic browning

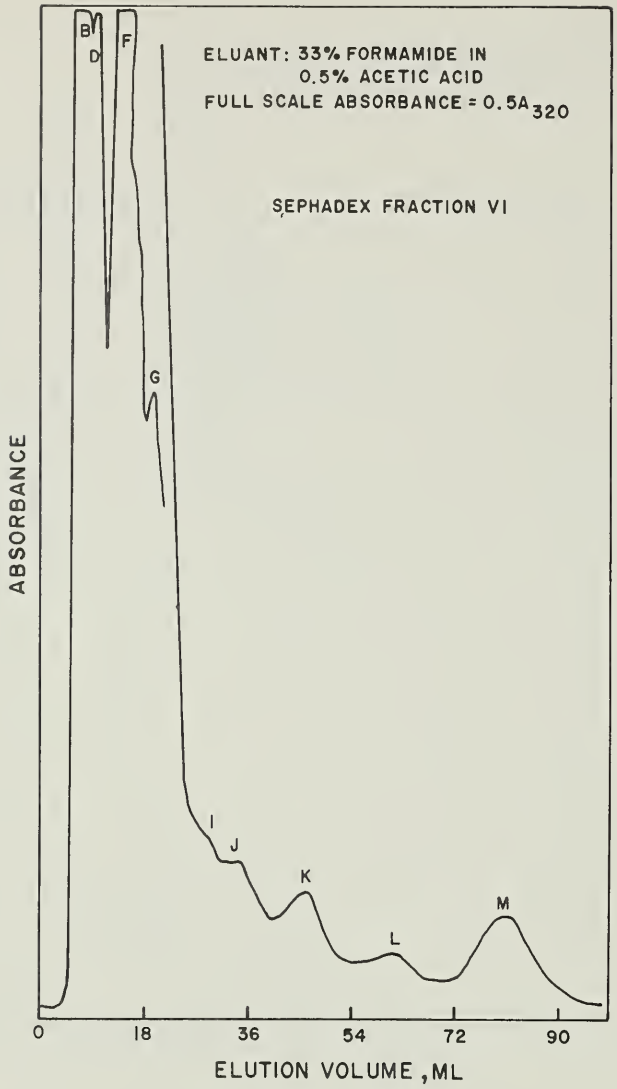

FIGURE 7--Fractionation of Sephadex fractions.

reactions produced high molecular weight colorants at the expense of low molecular weight naturally occurring pigments[4]. The results of HPLC fractionations on juice samples saved from these tests are shown in figure 12, which includes also a comparison with GSC.

About two dozen colorants can be seen in the first expressed juice. Of these, about five appear to be the main substrates for enzymatic browning. These are absent or significantly reduced in normal juice. Some of the original cane pigments (juice from heated cane) survive extraction conditions and are seen not only in the juice produced under normal conditions but are also found as components of the granulated sugar colorant. Earlier it was observed

[4]Smith, N. H. 1976. Inhibition of enzymatic browning in sugar processing. Int. Sugar J. 78: 259-263.

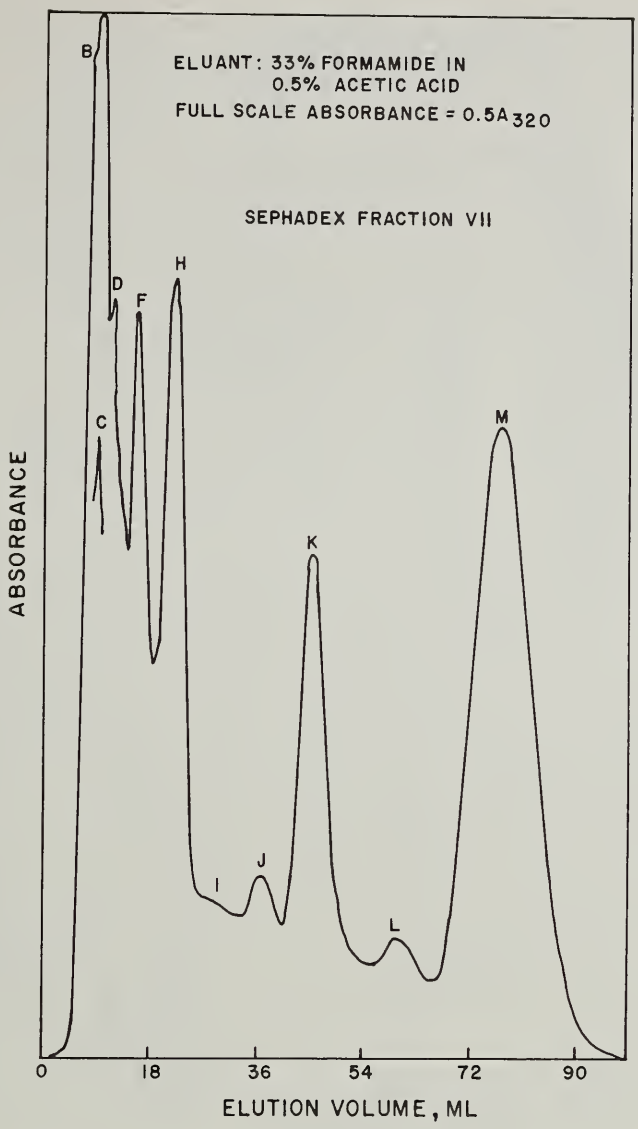

FIGURE 8--Fractionation of Sephadex fractions.

that the main colorants found in GSC are also found in raw sugar. This relationship is now extended to include raw juice as a source of substances directly contributing to granulated sugar color.

SUMMARY

Fractionation using the high pressure liquid chromatographic system described above has been found to be a versatile alternative to gel filtration. Not all of the parameters have been explored in detail, and it is expected that further development will yield more information than has been previously obtained concerning the origin and nature of sugar colorants.

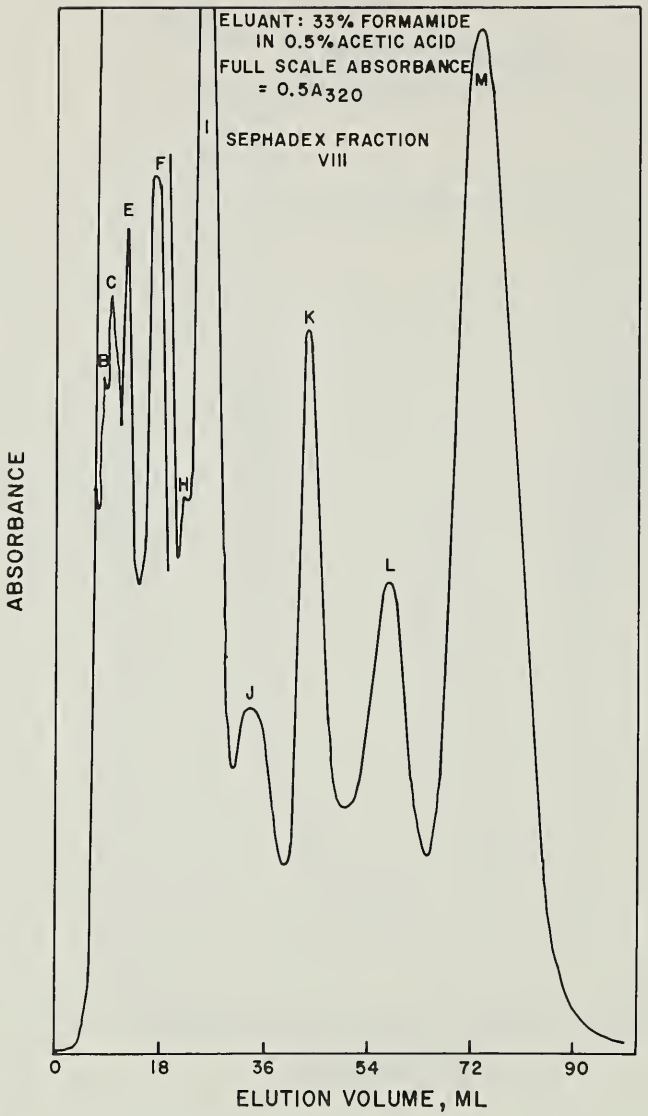

FIGURE 9--Fractionation of Sephadex fractions.

DISCUSSION

R. L. Knecht (Colonial Sugars): You showed fairly close agreement in your profiles with Hawaiian raw sources. Did you have a chance to profile any other raws, and if so, did you find a similar agreement?

N. H. Smith (C&H): I started that work, but at about that time I was beginning to have problems with columns plugging up. I couldn't tell whether differences were due to sources of sugar or to column problems.

R. L. Knecht: We have heard a lot about liquid chromatography. This is only the first of three papers today. Do you see it moving out of the research and applied research laboratories into the refinery control laboratories? Do you believe its ease of separating sugar, colorants, and other non-sugars make HPLC a useful tool for the day to day control of the processing of raw sugars into refined?

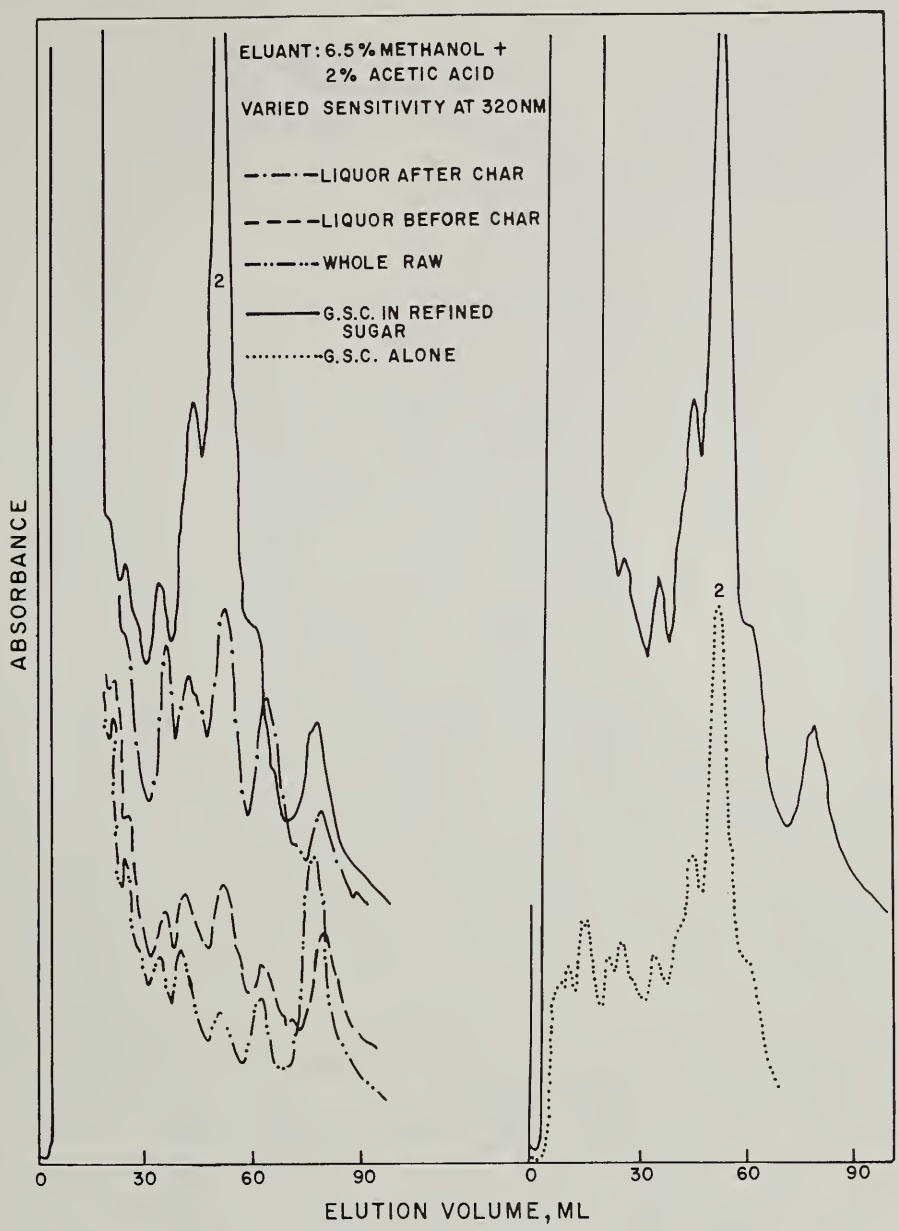

FIGURE 10--Fractionation of process streams.

N. H. Smith: It would be practical to use HPLC in the control laboratory, if we found a key component such as a colorant, the quantity of which correlated with sugar quality, and a method of removing that component preferentially.

K. R. Hanson (Amstar): You used organic solvents to establish a gradient. What would happen if you used a pH gradient?

N. H. Smith: I did try pH gradient, but because of the nature of the packing the pH did not change uniformly. Apparently, the column has an affinity for base so that the pH increase "broke through" like the end of a titration. It was a good idea because acidic colorants become ionic if the pH is raised and a separation should be achieved based on differences in acid strengths of the colorants. It would be an alternative method of separation which may work, using an appropriate column.

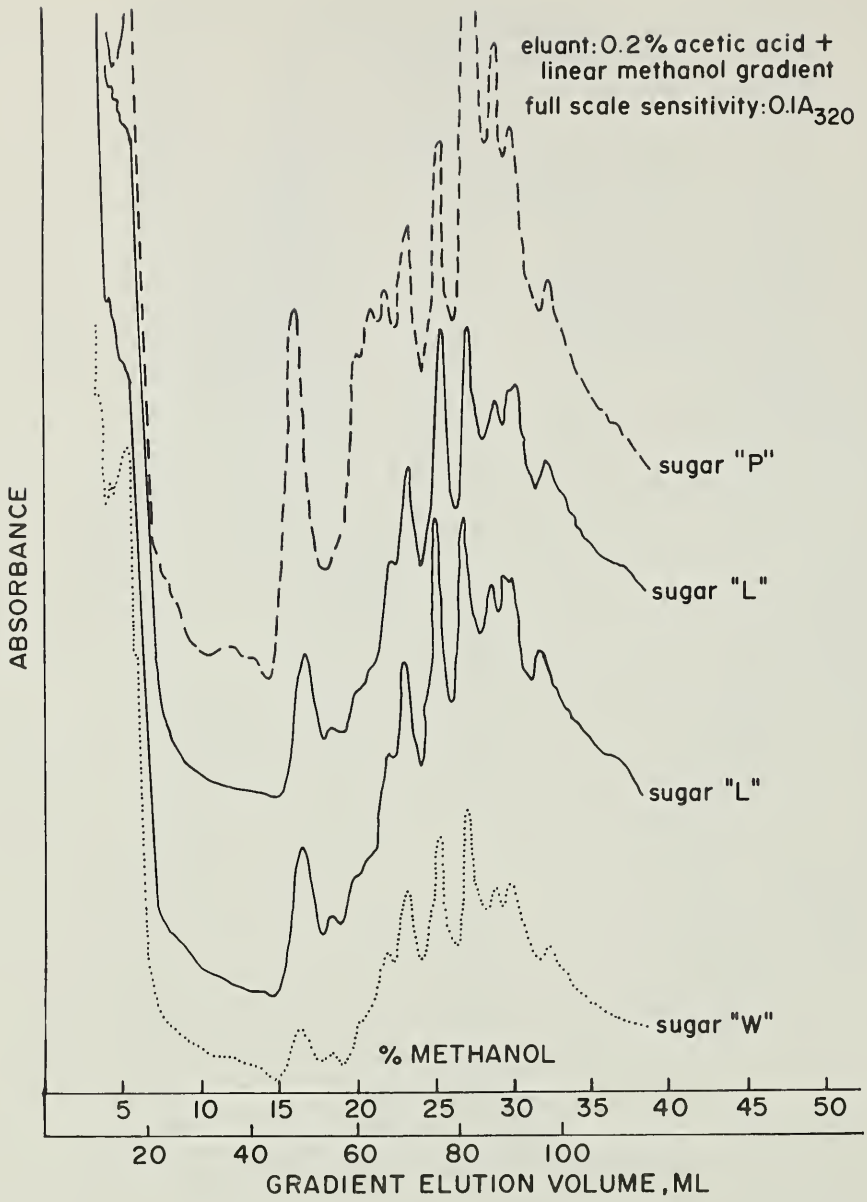

FIGURE 11--Fractionation of raw sugars.

H. G. Gerstner (Colonial): Can you weigh this colorant? How much colorant is that in granulated sugar by weight?

N. H. Smith: The sugar colorant was estimated as about 30 ppm in granulated sugar.

R. Cormier (Redpath): A U.V. Detector gives sensitive response to colorants but its response is not as proportional to the mass of the molecules as that of a R.I. detector. Such an isocratic system is usually very adequate for applications of R.I. Do you have any results using it?

N. H. Smith: I have not tried refractive index monitoring in sugar-free samples to see if I can detect the colorants. If the sample is predominantly sugar, the effect on refractive index is as bad as that using a solvent gradient, since the sugar is eluted before the solvents.

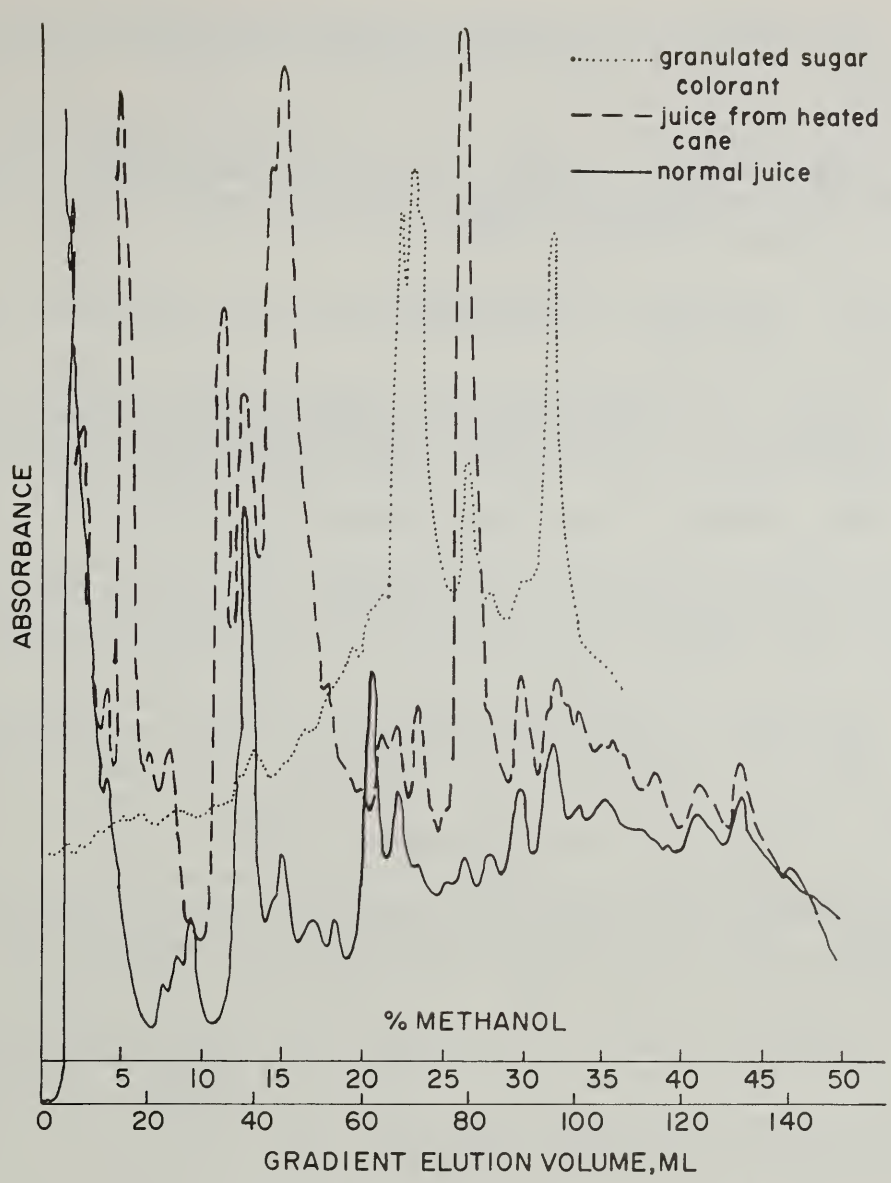

FIGURE 12--Fractionation of cane juice.

J. F. Dowling (Refined Syrups): Since you have the Waters HPLC system with recycling capacity, have you tried recycling the same peak repeatedly through the system to see if you can separate it into many peaks?

N. H. Smith: No, we haven't tried recycling colorants. We have tried recycling sugars, but in our system the recycling did not improve resolution. The peaks get broader and lower. Perhaps there is something wrong with our system, but the recycling did not give the desired improvement.

J. F. Dowling: Do you think you are getting decomposition?

N. H. Smith: No, I don't think it is decomposition. I just don't think the recycling technique worked for us as well as the Waters Company claims it does.

J. F. Dowling: Would not recycling have the possibility of separating one peak into several components that might possible be there?

N. H. Smith: Yes, that is the purpose of recycling. We do this in effect, by collecting a fraction and running it through again. This does seem to give an improvement in separation.

R. Cormier: Using a similar system, but with sucrose esters of fatty acids, we found that broadening of the peaks occurs to such an extent upon recycling that the practicability was impaired.

E. Obst (Amstar): How many of these peaks have you been able to identify as particular chemical compounds?

N. H. Smith: So far, we haven't tried to identify them; however, I have run through a few things such as chlorogenic acid, which came out corresponding to one particular peak. There are so many peaks that anything you put in is likely to match a peak. That is one of the problems.

J. B. Eldridge (Atlantic Sugar): Do you think that your method of isolating the colorants by ion exchange and methanol elution could have altered the colorants?

N. H. Smith: Actually it wasn't ion exchange - we used a nonionic resin as an absorbent. There is a possiblity of alteration. One peak in the granulated sugar colorant does not appear in any other sources of sugar. Generally speaking, almost all of the peaks found in isolated colorants also show up in other samples where the probability of chemical change is minimal: raw sugars, for instance. It is possible that chemical changes in processing can modify others which naturally occur, but I don't see evidence of new colorants that I have artifically made.

S. E. Bichsel (American Crystal): It appears that you have a much sharper tool than the old gel filtration method of isolating colorants. Will you go on and use this method to characterize various adsorbents: bone char, Synthad, granular carbon, and so on, as to color removal?

N. H. Smith: I would like to look at different sources of sugar to see if I can find variations in colorant composition, as well as follow the refining process to determine whether some color removing steps are selective.

R. S. Patterson (C&H): We seem to keep opening up new avenues of investigation faster than we can finish the old ones.

ANALYSIS OF FATTY ACID ESTERS OF SUCROSE BY MEANS OF HPLC

By Richard Cormier[1], L. H. Mai[1], and Philippe Pommez[2]

(Presented by Claude Gagnon[3])

ABSTRACT

The fatty acid esters of sucrose are "new" chemicals which contribute increasingly to the safety and comfort of human life. It has become important to have a fast and reliable method of analysis for their evaluation.

An analytical method based on high pressure liquid chromatography has been developed in order to separate and identify these sucrose fatty acid esters. The monoesters and diesters of sucrose stearate have been separated and quantitated and a complex mixture of sucrose tallowate has also been analyzed. The determinations were made using a one meter column packed with Fluoroether/Sil-X-I, with a solvent consisting of 91% n-chloropropane, and 9% anhydrous methanol. The peaks of positional isomers are then measured for each type of ester.

INTRODUCTION

Since the early sixties, when Lemieux[4,5] prepared and isolated sucrose monomyristate in a highly pure form, the preparation of fatty acid esters of sucrose has undergone many developments. They are no longer a laboratory curiosity, and have found in the recent past many industrial applications such as in detergents, and in cosmetics[6] formulations.

The degree of esterification is a very important parameter in the selection of the right sucrose esters. There is, therefore, a need for a fast and reliable method of analysis to show the amount and degree of esterification.

Current methods of analysis are based on thin layer chromatography and gas chromatography; the former is very long, tedious and suffers from some inaccuracy in the quantitation, while the later, although more practical, suffers

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[3]Research Supervisor, Redpath Sugars, Ltd., P.O. Box 490, Montreal, P.Q., Canada H3C 2T5

[4]Lemieux, R. V., and McInnes, A. G. 1962. Preparation of sucrose monoesters. Can. J. Chem. 40: 2376-2393

[5]_____. 1962. The composition of the sucrose monomyristate prepared by transesterification. Can. J. Chem. 40: 2394-2401

[6]Osipow, L., Marra, D., and Snell, F. D. 1957. Cosmetics containing sucrose esters. Drug and Cosmetic Industry 80(3): 312-313

from the need to prepare volatile derivatives, and from the difficulty in quantitating monoesters.

High pressure liquid chromatography (HPLC) does not require sample, pretreatments, and can allow the user to quantitate both mono- and diesters easily and rapidly.

INSTRUMENTS

The analyses were run on a Perkin-Elmer high pressure liquid chromatograph, model No. 1250, equipped with a U-V detector, model No. 250, and a refractive index (R.I.) detector fitted with a low prism suitable for work from 1.31 to 1.43 R.I. units.

Injections were made with a Hamilton syringe, model No. 710-1, into a 30 µl capacity loop injector. The columns used for the molecular exclusion and adsorption tests were, respectively, a Perkin Elmer VIT-X-628, 2.6 mm x 1 m, and a Perkin Elmer 2.6 mm x 1 m, packed with Fluoroether-Sil-X-1. The chromatograms were recorded on a Brinkman recorder, model "Servogor 2S", the peaks being quantitated by an Autolab System I electronic integrator. The required thin layer chromatograms (t.l.c.) were done on Brinkman silica gel "Sil-plate 22" coated plates, 20 cm x 20 cm.

ANALYTICAL PROCEDURES

The sucrose ester solutions were made by dissolving 2 g of the esters in 100 ml of a solvent prepared as follows: n-chloropropane:anhydrous methanol, 91%:9%. Solvent flow rate was 0.48 ml/min, and pressure drop across the column was 300 psi.

During the elution of the column, fractions of the effluent were manually collected according to the elution times of the various constituents as indicated by the peaks on the chromatogram. Those fractions were then concentrated and spotted on a t.l.c. plate using a mixture of toluene, ethyl acetate and ethanol in the volumetric ratio of 10:5:5 to elute the spots. A charring spray was then used to show up the spots.

RESULTS AND DISCUSSION

The following discussion will refer to sucrose tallowate unless otherwise specified, as it represents one of the most complex end mixtures. It is generally the result of a transesterification reaction between tallow (mainly triglyceride as shown below), and sucrose.

$$\begin{array}{l} CH_2\text{-}O\text{-}R_1 \\ | \\ CH\text{-}O\text{-}R_2 \\ | \\ CH_2\text{-}O\text{-}R_3 \end{array}$$

R_1, R_2, R_3 = H, or,

oleyl: $CH_3-(CH_2)_7-CH=CH-(CH_2)_7-\overset{O}{\overset{\|}{C}}-$, or,

stearyl: $CH_3-(CH_2)16-\overset{O}{\overset{\|}{C}}-$, or,

palmityl: $CH_3-(CH_2)_{14}-\overset{O}{\overset{\|}{C}}-$.

Tallow, being a natural product, has a composition which varies with its source and subsequent purification processes. However, the fatty acids radicals oleyl, stearyl, and palmityl represent the major constituents.

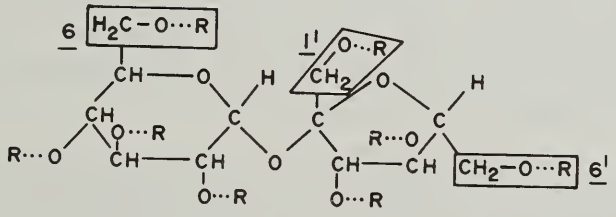

FIGURE 1--Structure of sucrose tallowate.

The molecule of sucrose tallowate itself is much more complex. From figure 1, it can be seen that there are eight possible sites in the sucrose molecule for substitution. Nevertheless, York et al.[7] have found the positions 6, 1', and 6' to be the preferred sites in the synthesis of sucrose monolaurate. Furthermore, Lemieux and McInnes[8] arrived at a similar conclusion for sucrose monomyristate. This reduces somewhat the number of possible constituents of the end mixture, although it must be realized that the final reaction product is a mixture of monoesters, diesters, triesters, or higher, together with the possible presence of residual glycerides; for instance, in one of our typical preparations, as many as eleven spots were detected on the t.l.c. plate. Obviously components for analysis must be defined.

It was decided to investigate more specifically the detection of the mono- and diesters. The molecular exclusion method was the first type of analysis considered but the figures in table 1 show that the molecular weights are so close as to prevent use of this method.

Turning then to conventional methods of separation, the chemical characteristics of the sucrose ester molecule must be considered. Being a surfactant,

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[7] York, W. C., Finchler, A., Osipon, L., and Snell, F. D. 1956. Structural studies on sucrose monolaurate. J. Am. Oil Chem. Soc. 33: 424-426

[8] Lemieux and McInnes, cited in footnote 5

TABLE 1--Molecular weights of some species

Sucrose esters	Mono-	di-	tri-	Triglycerides
Oleate	606	870	1134	884--olein
Stearate	608	874	1140	890--stearin
Palmitate	580	818	1056	806--palmitin

that molecule can be expected to have an ambivalent type of behavior because its long alkyl chain, non-polar and lipophilic, comprising the fatty acid moiety, is just the opposite of the polar, hydrophilic carbohydrate skeleton. Moreover, the expected impurities (the glycerides) do not have any pronounced polar moiety. Hence, adsorptive chromatography with a polar stationary phase should be suitable, as in Lemieux's work with silicic acid as the polar phase[9].

A procedure similar to Lemieux's cannot however be used because HPLC suffers a severe limitation as far as detectors are concerned. An R.I. detector is required to detect sucrose esters, but it does not allow solvent gradients, which Lemieux used. A new type of packing, less polar than silicic acid but polar enough to discriminate among the esters of interest, was then chosen. The stationary phase consisted of a liquid phase (Fluoro-ether), permanently bound to a silica type of support (Sil-X-I).

The solvent composition was a mixture of n-chloropropane and anhydrous methanol in a volumetric ratio of 91 to 9. It was noted that even small changes in the above composition greatly affect the aspect of the resulting chromatograms.

The figures 2 and 3 show, respectively, the chromatogram of commercial sucrose mono- and distearate samples. The numbering of the peaks refers to the numbered spots on the corresponding t.l.c. analysis (figs. 2A and 3A) made on the collected fractions. It is easy to see that both products have similar chromatograms, but with varying contributions from each peak. The identification of the peaks was done by comparison of their R_f's with those published by Gee[10] and by Bingham[11].

It was observed that the compounds are eluted in order of increasing polarity. It was found that peaks Nos. 6 and 7 correspond to the monoesters. The diesters appear as peaks No. 4 and No. 5; peak No. 3 corresponds to triesters. The situation becomes more complex in analysis of a sucrose tallowate sample, where peak No. 3 does not represent only the triesters, but also the residual monoglycerides (as shown by studies made with glycerides of oleic acid), while peak No. 2 is found to represent residual di- and triglycerides, as shown in figure 4.

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[9] Lemieux and McInnes, cited in footnotes 4 and 5

[10] Gee, M. 1962. Thin layer chromatography of sucrose esters and mixtures of raffinose and sucrose. J. Chromatog. 9: 278-282

[11] Bingham, S. B., and Kurtz, E. B., Jr. 1966. Arizona Agric. Expt. Sta., Tech. Bull. 176 pp. 18-22

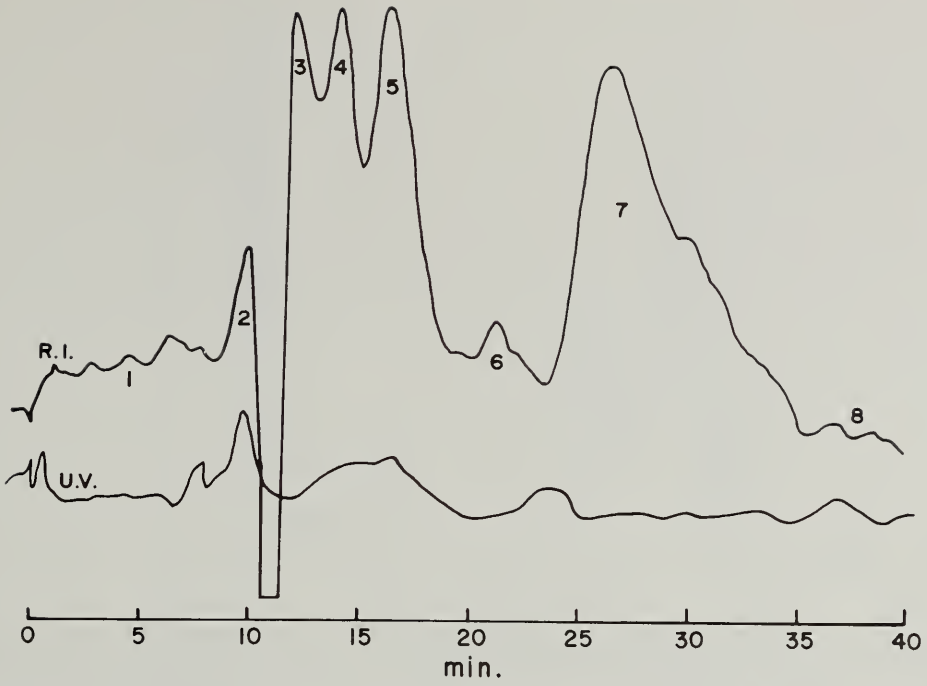

FIGURE 2--HPLC separation of commercial sucrose monostearate.

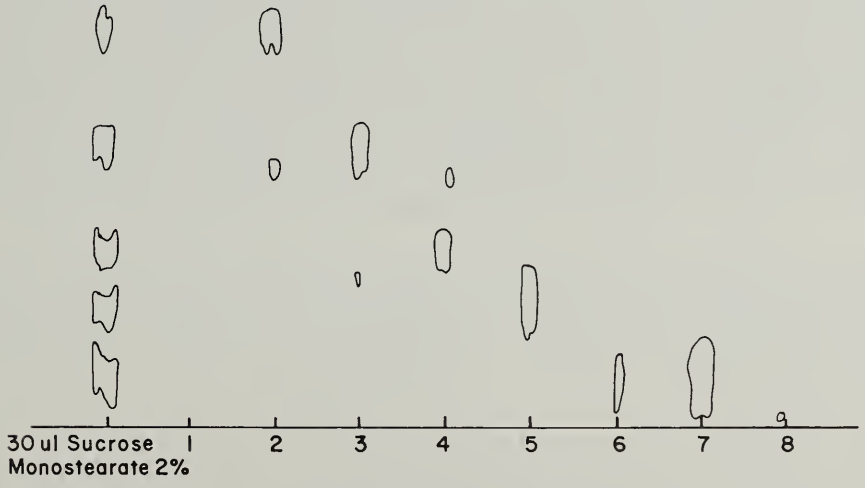

FIGURE 2A--TLC separation of commercial sucrose monostearate and fractions collected from HPLC.

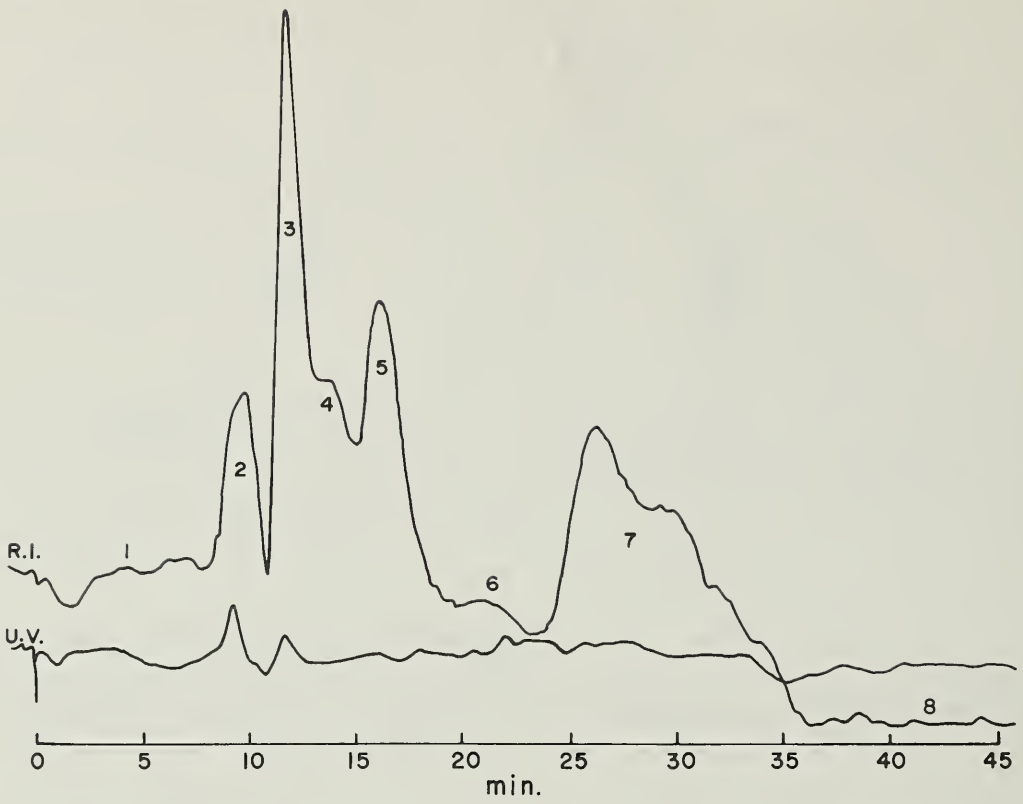

FIGURE 3--HPLC separation of commercial sucrose distearate.

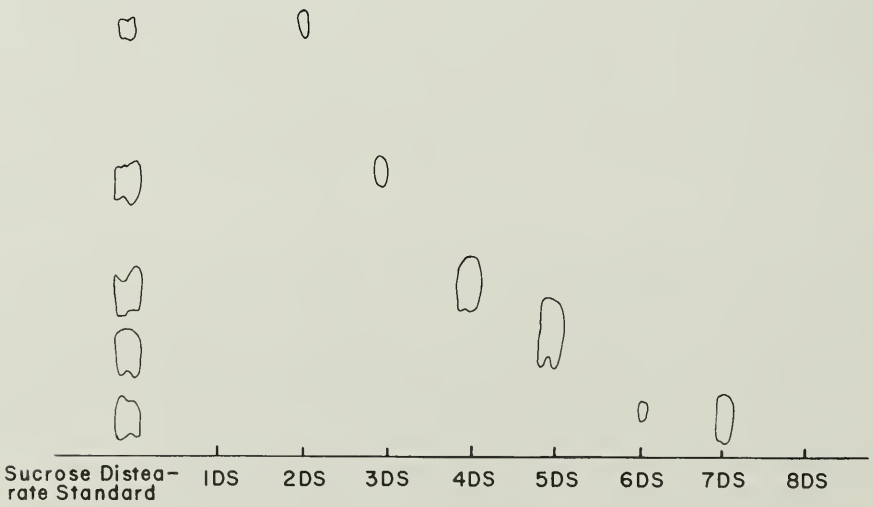

FIGURE 3A--TLC separation of commercial sucrose distearate and fractions collected from HPLC.

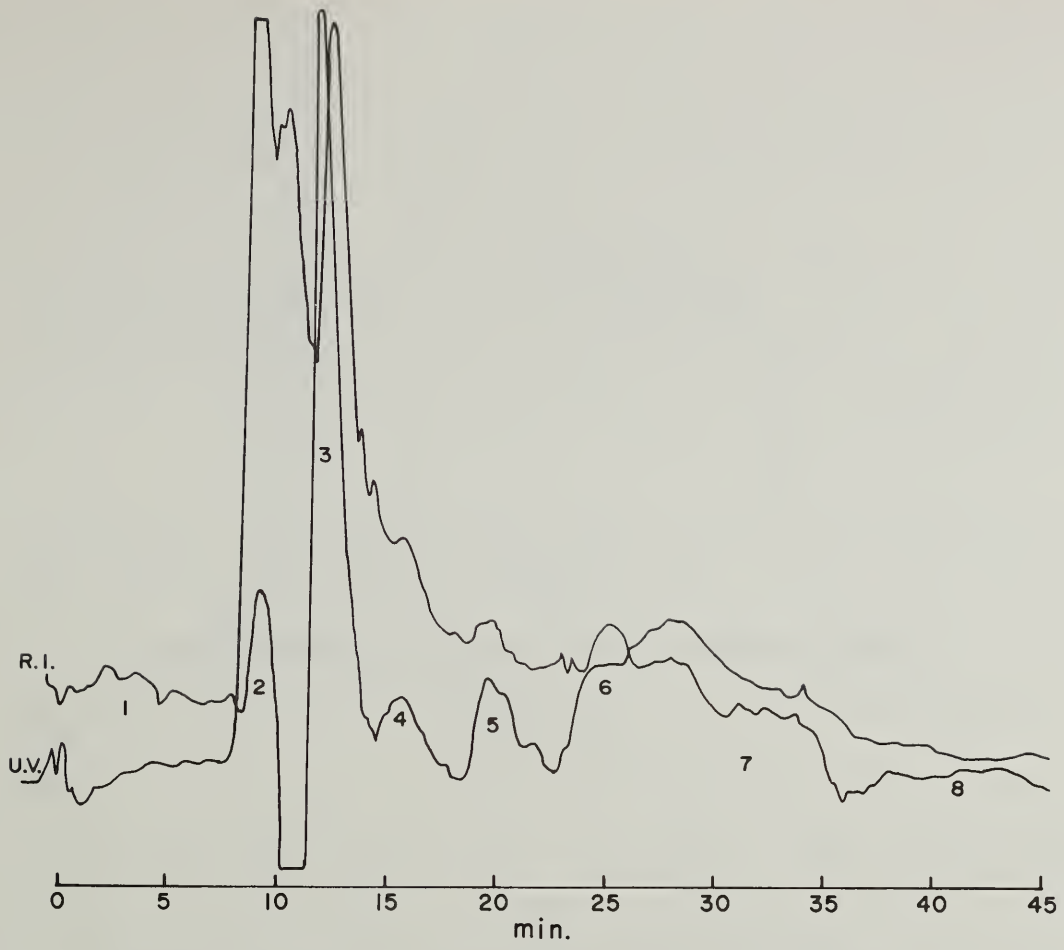

FIGURE 4--HPLC separation of commercial sucrose tallowate.

One of the serious problems encountered in the course of this work was the difficulty in obtaining pure sucrose esters according to their degree of esterification, in order to find the response coefficients for the type of detector used. But, although there is still a problem, recent developments in the production of sucrose esters has made pure compounds available[12]. Figures 5 and 6 show the chromatograms of pure mono- and ditallowate respectively; figure 7 shows a chromatogram of a commercial sucrose dioleate.

Assuming that mono- and diesters have identical response coefficients, some chromatograms were quantitated for their relative content in mono-, di-, and triesters with an electronic integrator. Table 2 reports the results obtained.

CONCLUSION

In conclusion, it is believed that this technique is a fast and reliable method of analysis for the fatty acid esters of sucrose, for their mono- and

[12]Talrès Development Ltd., Sterling House, 31/39 South St., Reading, England RG1 4QU

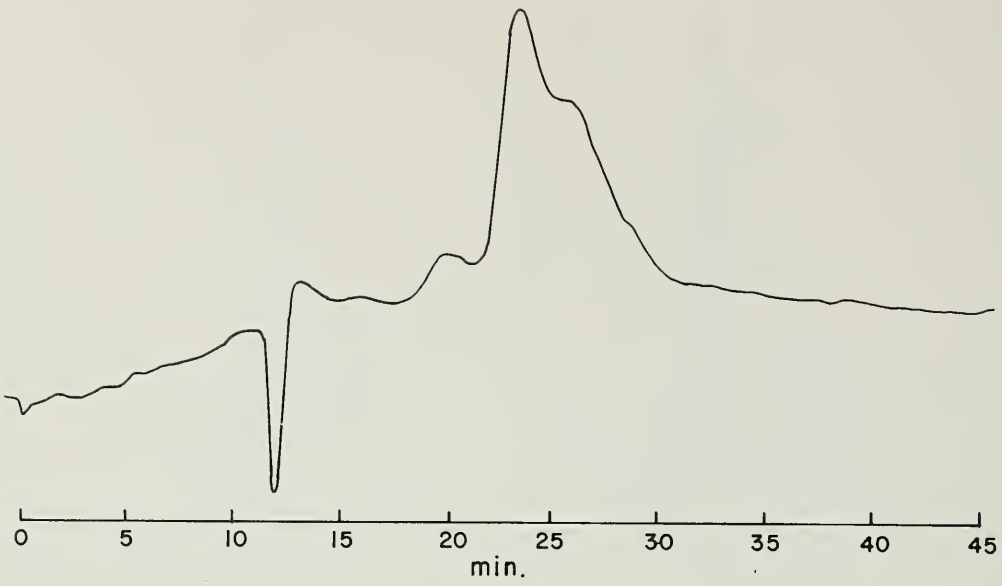

FIGURE 5---HPLC of pure sucrose monotallowate.

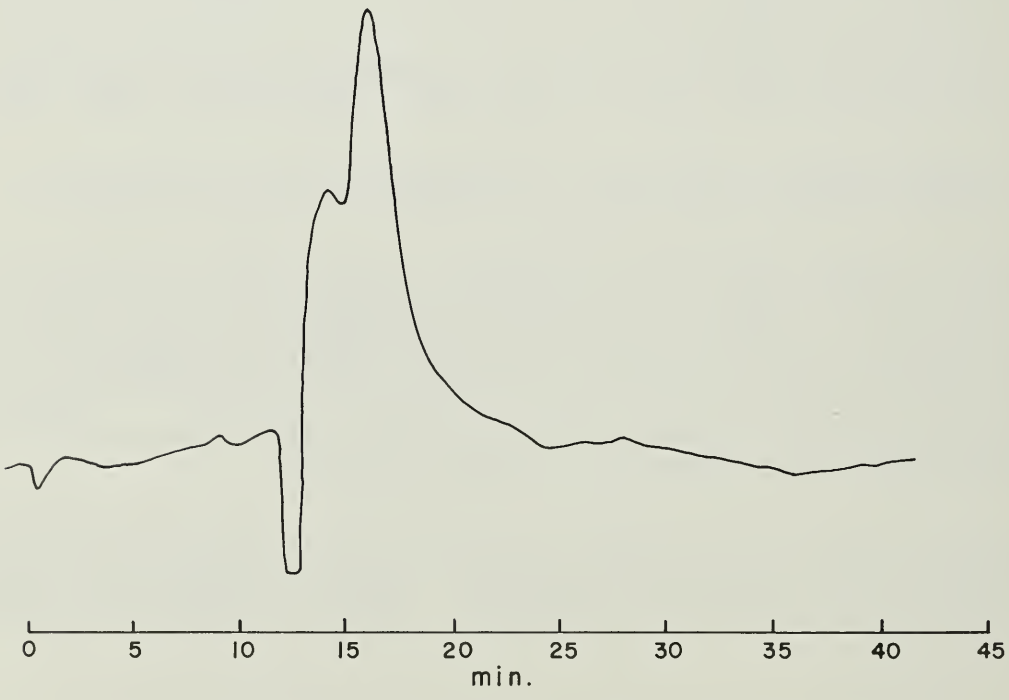

FIGURE 6--HPLC of pure sucrose ditallowate.

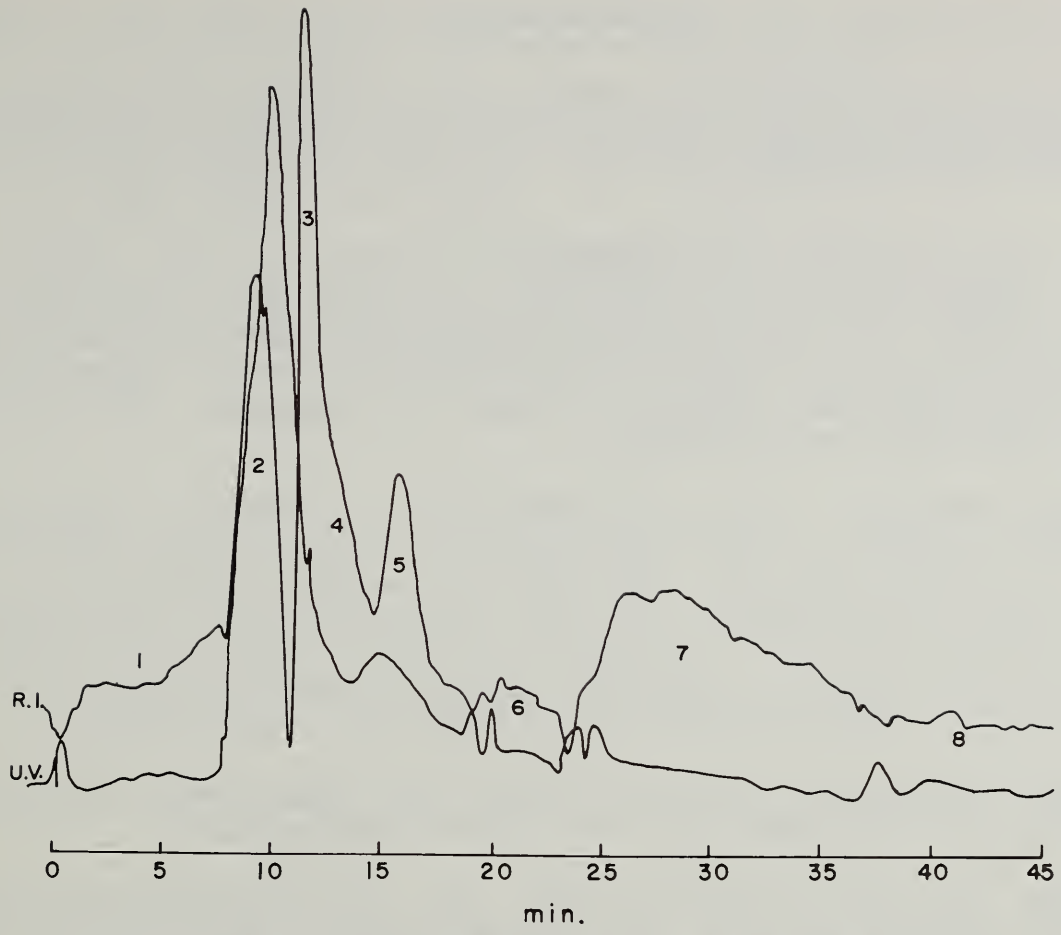

FIGURE 7--HPLC of commercial sucrose dioleate.

TABLE 2--Relative percentage of mono-, di-, and triesters

Sample	mono	di	tri
Sucrose monostearate	48.0	44.8	7.2
Sucrose distearate	38.3	33.2	28.5
"Talrès" monotallowate	98.1	1.9	-
"Talrès" ditallowate	5.1	94.9	-

diester contents. It is probable that minor modifications to the solvent composition would result in better resolution of the structural isomers of sucrose monoesters.

ACKNOWLEDGEMENT

We would like to thank Redpath Sugars, Ltd., for the authorization to publish, and also to thank Mr. P. Brion for his very valuable contribution to the study on sucrose tallowate.

DISCUSSION

M. Matic (SMRI): At the beginning, you mentioned that one of the reasons you are using high pressure liquid chromatography was that gas chromatography requires derivatization and, therefore, becomes less accurate. What is the precision of the method you are using?

C. Gagnon (Redpath): We are now working with an electronic integrator, but initially our integration was done manually. So, we had some difficulty in getting the desired precision. But with the electronic integrator and and without sample pretreatment - with only the dissolution of the sucrose esters in the solvent mixture - the reproducibility of each analysis is at about a tenth to two-tenths of a percent, and the accuracy is increased to its optimum which is still, however, in the order of a few percents.

M. Matic: All the peaks that you had were not single peaks. They seem to be made up of more than one peak. Are these stereoisomers?

C. Gagnon: Yes, that is what we think. For the monoesters, we probably have one major compound per peak, this compound being the ester of the primary alcohol at position 6 of the glucose moiety. But, for the di- and higher esters there are surely many compounds under each peak for there are so many combinations possible.

R. Cormier (Redpath): One comment: we did not really concentrate our efforts on a study of the accuracy but rather on finding a method that would rapidly, and without too much effort, separate sucrose monoesters from diesters.

Nevertheless we do believe that avoiding the step of sample pretreatment (or derivatization) should enhance the precision of the method.

C. Chou (Amstar): Can you give me some idea of the optimal reaction temperature and the rate of reaction? And, to what degree is the reaction involved for this esterification between sucrose and tallow?

C. Gagnon: We have primarily been concerned here with method of analysis. Most of the work concerning the esterification reaction itself has been done by Tate and Lyle, Inc., and all the details should be available in their patents concerning these sucrose esters.

N. H. Smith (C&H): Did you follow through on the purity of the compounds that you separated?

C. Gagnon: We believe that we got good purity of the compounds that we separated. Their purity was checked out by thin layer chromatography (t.l.c.).

It was observed that most of the compounds obtained after separation gave a clear unique spot, particularly in the case of the monoesters.

R. Cormier: We noticed, after peak No. 2 and before peak No. 3, a negative peak whose amplitude seemed to be somewhat related to the state of purity of the sample (namely its color).

N. H. Smith: With the refractive index detector, negative peaks usually mean that the index of the sample is on the other side of the solvent from those that you are getting positive peaks for.

A STUDY OF SUGAR INVERSION LOSSES BY HIGH PRESSURE LIQUID CHROMATOGRAPHY (HPLC)

By Margaret A. Clarke[1], Mary Ann Brannan[1], and Frank G. Carpenter[2]

(Presented by Margaret A. Clarke)

ABSTRACT

Decomposition of sucrose to glucose and fructose accounts for some sucrose loss in processing. HPLC is used to follow sucrose decomposition under typical refinery condition ranges of temperature, pH, and concentration.

INTRODUCTION

Sugar losses, known and unknown, result when sucrose breaks down under heat and other stresses in the refinery. Under acid conditons, sucrose inverts to glucose and fructose; under basic, sucrose decomposes to form, after many steps, lactic acid and other organic compounds including colorants.

Recent developments in high pressure liquid chromatography (HPLC) have made it a very suitable technique for sugar analysis.

Initial results from a study of sucrose breakdown under refinery conditions, using HPLC to determine sucrose, glucose, and fructose, are presented herein. It must be emphasized that these results are preliminary, and express current progress in the study. Several weaknesses in experimental technique in sample preparation were revealed; the study is to be redesigned using more accurate techniques, to obtain quantitative results.

Sucrose Destruction - Acid Conditions

$$\text{Sucrose} + H_2O \xrightarrow[\text{acid}]{\text{heat}} \underbrace{\text{Glucose} + \text{Fructose}}_{\text{invert sugar}}$$

This reaction has been studied extensively with regard to its mechanism and rate constant (2, 7, 11, 12)[3]. It is a second order reaction, dependent on concentrations of hydrogen ion and solvent and directly dependent on temperature.

There are two main problems in applying results from these studies to typical refinery conditions of high Brix and temperatures above 60°C. In most

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[3]Numbers in parentheses refer to items under "References" at the end of this paper

previous work, the rate of reaction has been traced by following the formation of the reaction products or the progress of a side reaction, e.g., change in pH, or by following the decrease in sucrose concentration. The reaction products have been analyzed by a copper-reducing sugars titration, which determines total reducing substances, i.e. the sum of glucose + fructose + interfering substances. Change in sucrose concentration has been observed by polarization or by determination of refractive index, both of which analyses are properties of the sum of substances in solution and give no indication of what products are formed. In no case have the concentrations of the specific sugars been measured.

The other problem is that much of this work has been done at low concentrations of sucrose, from 0.1 M (about 3 Brix) to 0.001 M. Under these conditions, water concentration is regarded as constant, but under refining conditions (60 Brix or 40% H_2O), water concentration is not constant. Ionic strength has also usually been regarded as a noninfluencing factor in very dilute solutions, which is not the case in more concentrated solutions.

Sucrose Destruction - Basic Conditions

Under basic conditions, sucrose decomposes into many compounds including colorants and color precursors. Again, progress of the reaction has been followed (1, 6, 8, 11) by observation of compounds formed, sometimes with paper chromatography which gives poor quantitation, or by measurement of the decrease in sucrose concentration (by refractometer) or by measurement of the amount of hydroxide ion added to maintain constant pH. In this last case, the hydroxide added is equivalent to the amount of hydroxide that has reacted with sucrose.

There has been much controversy about the initial steps in sucrose decomposition under basic conditions, particularly about the possible fromation of glucose and fructose. Current theory holds that a water molecule is pulled out of the fructose side of the sucrose molecule, breaking the glycosidic linkage, and allowing glucose to be split off (8). Thus glucose and fructose could appear as reaction products, although above pH 8 their formation would not be through the acid inversion mechanism. As with studies on acid destruction, most studies have been done in very dilute solutions. Because of the large number of reactions, rate observations are difficult, and the number of influential factors is high. It is known that various inorganic ions (e.g. Fe++) increase the rate of decomposition (14).

Athenstedt (1), a principal worker in this field, observed that decomposition occurs much more rapidly in beet juice than in laboratory solutions. Two possible explanations for this are that increased ionic strength increases activity of reactants, and that further reactions of intial reaction products will pull the initial reaction to completion. In this case, too, a means is required to observe, specifically, concentration of sucrose.

High Pressure Liquid Chromatography

The technique of HPLC, whereby components in solution are separated on a column of solid material, at high pressures and room temperature, appeared ideal for examining sugar solutions, for several reasons (3, 5, 9, 10). It is possible to inject samples directly in water solutions; no derivatization or pretreatment,

other than Millipore-type filtration, is required. A determination for glucose, fructose, and sucrose requires less than 15 minutes. HPLC separates sucrose, glucose, and fructose clearly, showing one peak for each sugar, with good separation for glucose and fructose. Detection is by refractive index change. There is no interference from other sugars or other substances likely to be in refinery liquors. If other sugars are present, they are shown on the chromatogram. With a column that is specific for carbohydrates, colorants and other organic compounds are not separated, and so do not appear on the chromatogram.

EXPERIMENTAL

A Waters Liquid Chromatograph, with Universal L. C. injector Model U6K, with Model 440 absorbance detector and Series R-400 differential refractometer, with Waters Micro-Bondapak-Carbohydrate column, was used to determine sugars. Conditions were: flow rate 1.5 ml/min, attenuation 8x. The solvent was acetonitrile - water, 83:17. Chromatograms were displayed on a Sargent Model MR recorder. The refractometer was the detector.

Samples were run at room temperature (20°C), at 40°C and 60°C in constant temperature water baths and at 80°C in a constant temperature oil bath. All samples were prepared at 60 Brix concentration, from a very high-grade, first-strike sugar. Samples at pH 2, 3, 4, 6, 8, and 10 were run over periods of one to twenty days. Ionic strength was varied from extremely low, in deionized water, to 0.1 M or 0.5 M adjusted with KCl. The 60 Brix samples were diluted 25:1 with distilled water, and filtered through 0.45 μ Millipore Filters, using a Swinney adaptor syringe, before injection. The solvent was also filtered through a 0.45 μ filter, and sonicated to remove air bubbles. Burdick and Jackson "distilled in glass" solvents were used. Peak heights were measured, and concentrations read from calibration curves prepared with standard solutions of glucose, fructose and sucrose. Samples, usually of 25 μl, were injected by Pressure-Lok liquid syringes (Precision Sampling Co.).

RESULTS AND DISCUSSION

The calibration curves were linear over a range of 0.0 to 0.4 mg for solutions from 20 to 100 mg/ml, but deviated from linear when range exceeded 0.0 to 0.2 mg for dilute solutions (<10 mg/ml). The curve closest to sample concentration was used for each sample sugar solution.

As expected, inversion rate varied over time with temperature and pH, as shown in figures 1, 2, and 3 for runs at pH 3 and 4 at 80°C and 60°C. In figure 1, the reaction is followed by observing decrease in sucrose concentration, and in figure 2, by observing increase in glucose concentration. In each case, the percent decrease or increase is calculated on the basis of amount of sucrose in the sample solution as prepared, and amounts of glucose and fructose possible if this sucrose were converted completely to glucose and fructose, with no other reactions. Table 1 shows results for runs at 80°C, pH 3.

A dependence on ionic strength was also observed: the reaction progressed more rapidly at higher ionic strengths, as shown in figures 1 and 2. Some workers have reported that inversion is not affected by change in ionic strength;

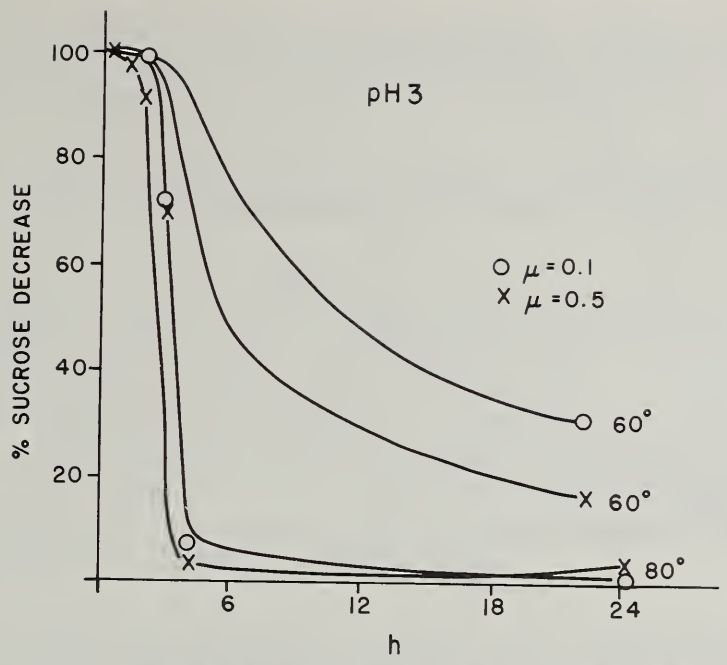

FIGURE 1--Decrease in sucrose concentration at 60° and 80°C, pH3, over 24 hours.

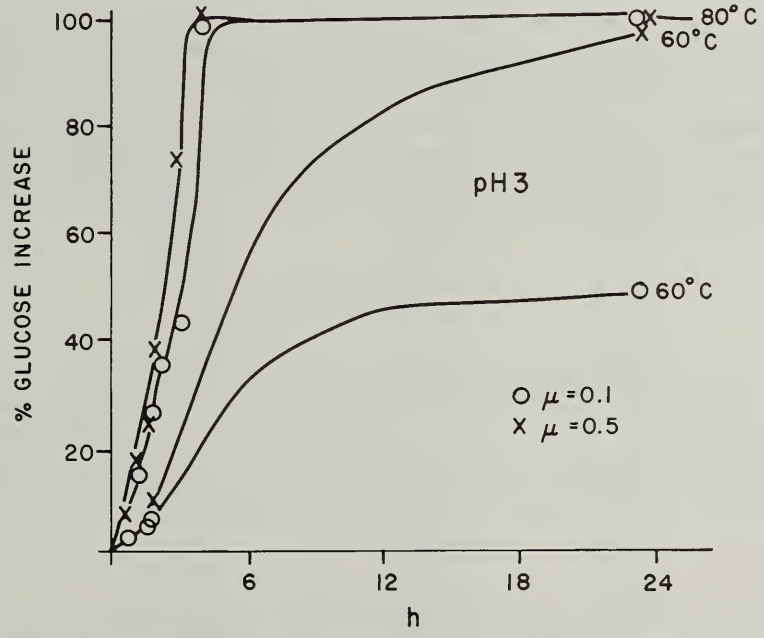

FIGURE 2--Increase in glucose concentration at 80°C, pH3, over 24 hours

while this may be a suitable approximation for dilute sugar solutions, it is obviously not so for the 60 Brix case. In other work, salt effects have been shown even at low concentrations (12).

Since the reaction is second order and dependent on solvent concentration, in this case water concentration, any factor affecting the activity of water, e.g., ionic strength, will affect the rate of reaction. As ionic strength

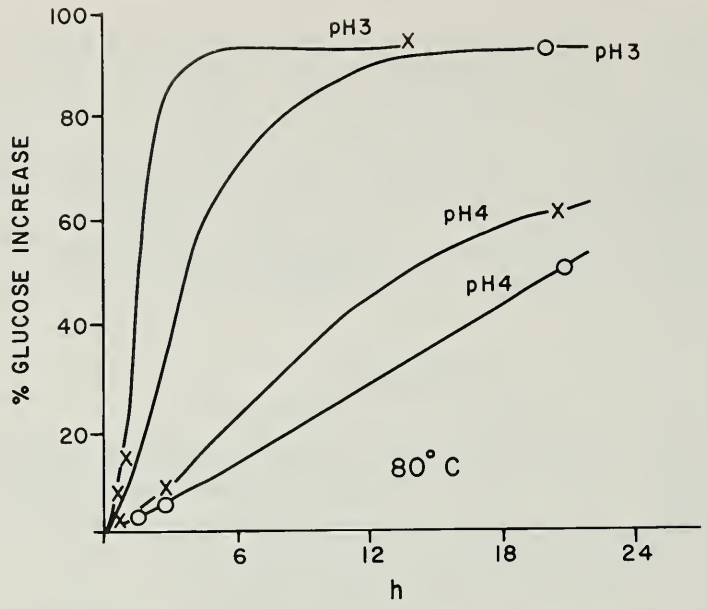

FIGURE 3--Increase in glucose concentration at pH 3 and pH 4, at 80%, over 24 hours. (0 μ = 0.1, X μ = 0.5)

TABLE 1--Sucrose inversion at 80°C, pH 3

	t(h)	Fructose* %	Glucose* %	Sucrose+ %
Ionic strength = 0.1 m/l	0.5	3.71	3.71	100.00
	1.0	12.38	14.86	100.00
	1.5	21.04	26.00	100.00
	2.0	27.23	35.90	99.00
	3.0	39.36	43.33	71.67
	4.0	40.36	99.03	7.69
	24.0	100.00	100.00	1.50
Ionic strength = 0.5	0.5	4.70	7.43	100.00
	1.0	14.86	17.33	100.00
	1.5	21.04	24.76	97.73
	2.0	28.47	37.88	91.22
	3.0	43.33	74.28	70.11
	4.0	100.00	100.00	3.26
	24.0	100.00	100.00	2.61

*Percent formed of total possible with complete inversion
+Percent of original concentration of sucrose

of 0.5 M the activity coefficient, and therefore the activity, of water are higher than at ionic strength of 0.1 M, and so the reaction rate increases because of its direct dependence on solvent concentration.

Figure 3 shows the influence of pH and ionic strength at constant temperature. Below 30°C, little decomposition is observed in the first 24 hr even at pH 3.

It was also observed that, with increasing time and particularly at low pH, fructose concentration became less than glucose concentration. Fructose undergoes further reactions more rapidly than does glucose. It was decided, therefore, to follow the formation of glucose as the most accurate indication of inversion progress. The observation about concentration difference emphasizes the error in the use of reducing titrations for total invert determination to follow the inversion reaction: total reducing substance in these cases is not made up of equal amounts of glucose and fructose.

Results for sucrose decomposition at basic pH's are not discussed here because results following disappearance of sucrose by HPLC do not have sufficient accuracy at this time. The appearance of glucose has greater accuracy because the glucose peak in HPLC is larger than the sucrose, for equal concentrations. It should be noted, however, that glucose and fructose are observed, along with the formation of colored substances, after treatment of basic 60 Brix sucrose solution with heat.

Because of problems in experimental technique (evaporation, sample loss) the accuracy of the figures in table 1 is poor. Precision of the measurement is very good, with a standard deviation of ± 0.01 for each of the three sugars over 20 measurements on standard solutions. Sample treatment procedures will be adjusted and the measurements repeated for greater accuracy.

SUMMARY

High pressure liquid chromatography is a suitable method to study sucrose inversion of typical refinery liquors and also to follow sucrose decomposition at basic pH. Experimental techniques must be further refined to increase accuracy. Results from the HPLC technique applied to typical refinery liquors will be useful in determination of sucrose losses.

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DISCUSSION

R. S. Patterson (C&H): I have the perennial question. You have studied here the inversion and decomposition of sucrose. Have you attempted any mass balance at all?

M. A. Clarke (CSRRP): No.

W. M. Nicol (Tate & Lyle): I find this paper a very relevant subject indeed for sugar refiners. The question is: at 80°C and a pH of 3 the rate of inversion is something like 4% or 5% per 15 min. How does this affect the measurement using your high pressure liquid chromatography?

M. A. Clarke: I have not compared our numbers with the literature yet. As soon as our sample reaction time is complete, the sample is frozen to stop further inversion.

W. M. Nicol: The question I would like to put is, how does the ionic strength relate to ash? This is rather important.

M. A. Clarke: The ionic strength of 0.1 m/l is about the level of ash in a raw sugar liquor.

W. M. Nicol: Was the pH measured at room temperature?

M. A. Clarke: Yes.

R. L. Knecht (Colonial): You mentioned that ions affect the decomposition of sucrose. How about the calcium and phosphate ions? Do you have any information on these?

M. A. Clarke: From one point of view there is the ionic strength effect and all the ions contribute to that; but, on the other hand, the individual ions - calcium, phosphate, etc. - can change the rate by specific reactions with the components of the system. There are two contributing factors to consider.

J. F. Dowling (Refined Syrups): We don't intentionally decompose sucrose. We may invert it, but this is not really decomposed.

M. A. Clarke: We say "decomposition" to include the reaction at basic pH as well, in which products other than invert are formed. Invert itself will decompose to other compounds under most conditions.

J. F. Dowling: If we are going to measure the dextrose-levulose changes for say char liquor, the level is perhaps 0.1%; there is a whole lot of sucrose there. Can you get the accuracy you need in this case?

M. A. Clarke: We now make two runs, one to analyze for sucrose, and one to analyze for the reducing sugars with different dilutions. This increases the accuracy of measurement when the concentration of one species is much greater than that of the other.

J. F. Dowling: We were unable to get that accuracy with gas chromatography. You could also run pure levulose and pure dextrose at low pH and high temperature to see if there is a new peak - I'm sure there is a new peak. In our gas chromatography work we found difructose dianhydride under sucrose.

M. A. Clarke: That may have been a result of the pretreatment required for gas chromatography. HPLC separates compounds on an entirely different basis than does GLC.

J. F. Dowling: It was a result of high pH and high temperature, but the peak was lost under sucrose. It is true that liquid chromatography with refractive index detection is a completely different system but there is a possibility that peaks can be hidden under one another such as with sucrose in GLC.

D. H. Foster (Sugar Research Institute, Queensland): I would like to ask if there is any sign of mannose in the reaction products at all. When we held syrups of 89 purity at 75°C for up to 48 hours, the pH fell from 6.0 to 5.4 and throughout this time mannose increased from 0.008% to 0.036% of the syrup.

M. A. Clarke: We didn't notice mannose and I don't think we'd get mannose under the conditions in which we are working, because we used very high purity

sugar to start with. If other sugars had been formed, I think we would have noticed them. Mannose can be a byproduct of the Leuconostoc microorganism under some conditions.

Another explanation for the presence of mannose is that, in heated sugar solutions, after liming, there is rearrangement of the glucose and fructose to mannose and psicose[1]. All four sugars are in an equilibrium that favors glucose and fructose.

D. H. Foster: Mannose concentration was increasing relative to glucose and fructose, but it was not increasing very rapidly.

N. H. Smith (C&H): You showed that the fructose didn't increase as rapidly as the glucose, indicating that the fructose was being converted to something else. You can't tell whether or not glucose is also degraded. If you run the simple sugars by themselves under the same conditions you can follow both fructose and glucose decomposition.

M. A. Clarke: That is a very good suggestion.

J. V. Lopez-Ona (National): I want to congratulate you on your paper - I really enjoyed it. I am always very much interested in sugar inversion losses. I also want to say that you are getting involved in a pretty complicated subject, because from what I heard you are playing around with 4 factors: temperature, pH, time, and ionic strength. You are also playing with levels for each of those factors. If you take 3 factors and you get 3 levels, you've got 27 combinations. If you go to 4 factors and 3 levels, you're going to have 4x4x4, or 64 combinations. About what you are going to measure, I don't have any questions; but, you've got a beautiful situation for a factorial analysis. I offer you help in designing this experiment. Since you have been working on this for only three months, that's what I call a pre-experiment. You now have enough information to look at the experimental design. We can decide whether to go into a full factorial or fractional factorial design. Without such a design, you might find yourself having to take 264 combinations and to try to make a reasonable conclusion about that would be a little difficult. I'll send you all the information which we worked out about two years ago about fractional factorial experiments.

M. A. Clarke: Thank you very much, John. We will take you up on that. I have been thinking that there is enough work here to last us for several years to come, and assistance in experimental design would be very useful.

M. Matic (SMRI): From your graphs, it appears that the rate of reaction has changed. There is a much faster rate at the beginning and then it levels off. Is this something that is normal and expected?

M. A. Clarke: I think that is because the change in percent is graphed, rather than an absolute measurement.

M. Matic: It is generally considered that fructose is the more reactive molecule. Yet, there is a lot of evidence that molasses, which is the end

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[1]Binkley, W. W., Roberts, E. J., Jackson, J. T., and Martin, L. F. 1963. The fate of cane juice simple sugars during molasses formation. Int. Sugar J. 65:169-173.

product of all the reactions contains, more fructose than glucose. This does not seem to agree with what your results suggest.

M. A. Clarke: We were not working with a complex mixture like molasses, but with a very pure refined sugar with some potassium chloride added. There are many reactions possible in molasses that could not occur in our system.

S. E. Bichsel (American Crystal): To amplify on what some of these gentlemen have said, we have a very complex situation here. Under hot alkaline conditions you've got sugar supposedly going to glucose and fructose, and, of course, fructose being very thermally labile breaks down, as reported in the literature, to lactic, acetic, formic, and saccharinic acids; and color bodies. Glucose supposedly does the same thing, only at a little slower rate. Under extremely hot acid conditions you have the formation of glucose and fructose; fructose goes on to hydroxymethylfurfural, and glucose is again less thermally labile. My question is, do you really feel that you can make a balance without taking into consideration the other breakdown products of glucose and fructose under alkaline and acidic conditions?

M. A. Clarke: We have not considered making a balance under the basic condition. The acidic condition is what we are going to work with, first and I think we will have a better chance at that.

S. E. Bichsel: I'd like to confirm your observations concerning rate of inversion with respect to ionic strength. We found, in some of our work producing liquid sugar directly from thick juice using deionization, we came out with conductivity ash down as low as 0.003%, whereas regular liquid sugar might be up around 0.015%. The inversion rate of the low ash liquid sugar, all other things being equal, is about twice what it is at the higher ash level.

K. H. Schoenrock (Amalgamated): I understand the quantitation of the monosaccharides was not as precise as you would like it.

M. A. Clarke: It was quite precise but it wasn't as accurate as we would have liked it to be. We are revising our procedures to increase accuracy.

K. H. Schoenrock: Did you control the temperature of the column or was it exposed to room temperature?

M. A. Clarke: It was at room temperature, which is somewhat controlled in our laboratory.

K. H. Schoenrock: We found in some of our preliminary work that the lack of temperature control many produce some of the background noise that you often see, such as in the slides in the first paper this morning. By controlling the temperature to within 0.01 degree, we've been able to establish a much better baseline.

M. A. Clarke: Most of our baseline trouble has just been with air, or with an insufficiently flushed reference compartment. Otherwise we have a very good base line.

K. H. Schoenrock: I wonder if anyone else has observed that column temperature control is critical for very precise work.

N. H. Smith: I have not done any work on the column temperature effect, but I do know that the cost goes up rapidly when you start getting close temperature control in chromatography. And of course, liquid chromatography isn't cheap to start with.

DETERMINATION OF REFINED SUGAR PROTEINS BY POLYACRYLAMIDE GEL ELECTROPHORESIS

By Mary An Godshall and Earl J. Roberts[1]

(Presented by Mary An Godshall)

ABSTRACT

Polyacrylamide gel electrophoresis was found to be an excellent method of separating protein from the protein-polysaccharide complex that is implicated as a major factor in acid beverage floc. The protein in several raw and refined sugars was examined by this method. Progress in quantification of this protein is also reported.

INTRODUCTION

The protein content of refinery products has not received as much attention as many other constituents, such as ash, invert, color, and polysaccharides, probably because the majority of the protein is removed during juice clarification, and most of the small amount remaining is removed during further stages in the process. It is, therefore, general belief that not enough protein remains in refined sugar to be a problem. Recently, however, there has been evidence linking protein to the mechanism of acid beverage floc formation.

Roberts and Martin (16)[2] analyzed the protein in the nondialyzable fraction of cane juice and found that the total protein in 23 of 30 juices constituted 0.5% to 0.7% of the juice on the basis of dry solids (5000 ppm to 7000 ppm protein). The amount of protein in refined sugars is extremely small, and few figures are cited in the literature. Ambler and Byall (1) analyzed white granulated sugars for protein nitrogen by tannic acid precipitation followed by Kjeldahl determination and, in 32 samples, found a protein nitrogen range of 0.0 ppm to 8.4 ppm. Earland and Ramsden (7) found that proteins are normally present in refined sugars in varying concentrations, but based their conclusions on visual observation of stained proteins after electrophoresis and did not quantitate their findings.

Roberts (13) reported that isolated acid beverage floc contained 0.63% nitrogen. If it is assumed that all the nitrogen came from protein, this can be interpreted as 4% protein. Cohen, et al, (5) observed that all acidified refined sugar solutions will floc if protein is present and theorized that acid floc arises from the aggregation of macromolecules in the form of a protein-polysaccharide complex. They felt that protein was the initiating component in floc formation. Stansbury and Hoffpauir (18) reported proteins in five acid floc-positive sugars, and they found that the protein averaged about 5% of the total floc.

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[2] Numbers in parentheses refer to items under "References" at the end of this paper.

With protein implicated as a possible major factor in floc formation, it became necessary to develop a method to isolate and analyze the protein fraction. An ideal method of measuring small amounts of protein is gel electrophoresis because of its sensitivity in the microgram range. It is adaptable to the investigation of the protein in virtually any biological material, and has, in fact, been used in the sugarcane industry to determine genetic variations (17, 19, 20) and isoenzymes in the cane plant (11). It has the added advantage that individual proteins can be directly measured as proteins instead of estimated from a nitrogen determination with the assumption that all nitrogen comes from protein.

Theory of Gel Electrophoresis

Gel electrophoresis is a separation technique that involves the migration of charged particles in an electric field through a porous but rigid electrophoresis medium. Separation of the charged molecules occurs because of their differing sizes, shapes, and charges under the experimental conditions. Because of the amphoteric nature of proteins, a variety of buffers of differing pH values can be used to manipulate the charge on the protein to control the direction of migration.

The electrophoretic medium consists of polyacrylamide gel, which is formed inside glass tubes from a mixture of the monomer acrylamide and a crosslinking agent such as N, N'-methylene bis-acrylamide. Polymerization is effected by a variety of oxidative procedures to give a firm, transparent gel. As an alternative, photopolymerization can be initiated in the presence of an activator such as riboflavin (12).

The concentration of acrylamide in the gel determines the stiffness or porosity of the gel and can be varied to suit the type of protein being analyzed. Usually two gels are used. The separating gel is the medium in which the separation takes place, and it occupies the majority of the electrophoresis tube. On top of this is situated a shorter, more porous stacking or spacer gel, which concentrates the sample proteins into narrow starting zones before they enter the separating gel. This step greatly improves the sharpness and reproducibility of the separation (6).

After electrophoresis, the gels are removed from the glass tubes and placed in a solution containing a fixative and a protein dye. Excess dye is removed from the background gel with an appropriate solvent, leaving the stained protein bands clearly visible.

EXPERIMENTAL

Isolation of Protein

Protein was separated from the sugars by dialysis. An appropriate weight of sample, usually 100 g to 200 g of solids, was subjected to dialysis in regenerated cellulose tubing (molecular weight cut-off of 12,000) for about 108 hours (4-1/2 days). Microbiological contamination was inhibited by dialyzing the samples against toluene-saturated distilled water.

After dialysis was completed, the nondialyzable material remaining in the dialysis bag was concentrated under reduced pressure to about 5 ml to 10 ml volume, frozen, and lyophilyzed.

No further purification of the protein was carried out prior to electrophoresis.

Preparation of the Gels

The gels were photopolymerized inside glass tubes of 1 mm wall thickness, 5 mm i.d. Two lengths were used: 7.5 cm tubes for the ribonuclease standard curve, and 10 cm tubes for determining protein content of the sugar samples. Table 1 gives the lengths of the components of each gel tube.

TABLE 1. Dimensions of gels inside electrophoresis tubes (cm)

7.5 cm length		
separating gel	5.5	
stacking gel	1.0	
sample	1.0	(capacity = 0.2 ml)
10 cm length		
separating gel	6.0	
stacking gel	1.0	
sample	3.0	(capacity = 0.6 ml)

Tubes were washed in detergent, thoroughly rinsed and soaked about 30 min in water containing a few drops of Triton X-100, and dried. (Use of Triton X-100 facilitated the removal of the gels from the tubes after electrophoresis.)

The bottom of the tubes were temporarily sealed with a paraffin film (Parafilm) and set into a supporting rack. The gel solutions were made up immediately before using, but the stock solutions were kept indefinitely in the refrigerator. All solutions were brought to room temperature prior to using.

The formulations for the gel stock solutions are listed in Table 2. The solution for making a 10% separating gel consisted of one part solution A, one part B, and two parts C. It was carefully pipetted into the tubes, a drop of water was layered on top to flatten the meniscus, and the filled tubes were placed in front of a 12W Cool-White fluorescent light (strong 365 nm emission) for 15 min to polymerize.

The water layer was removed from the polymerized gel with a wick of tissue and the stacking gel solution layered on top. The formulation for this gel consisted of one part each of solutions D, E, and F and four parts distilled water. The meniscus was again flattened out with a water drop and the gel polymerized in front of the light for 45 min. Figure 1 is a diagram of a glass electrophoresis tube containing the gels and sample.

Preparation of Sample for Electrophoresis

The sample to be analyzed was weighed into a 2 ml volumetric flask and brought up to volume with the same buffer to be used during electrophoresis. A

TABLE 2. Gel stock solutions

Solution A		Solution B	
Acrylamide	40 g	1 N HCL	24 ml
Bisacrylamide*	0.8 g	Tris+	18.1 g
Water to 100 ml: Filter		Temed±	0.12 ml
		Water to 100 ml	

Solution C		Solution D	
Riboflavin	1 mg	1 N HCL	48 ml
Water to 100 ml		Tris	5.98 g
		Water to 100 ml	

Solution E		Solution F	
Acrylamide	28 g	Riboflavin	4 mg
Bisacrylamide	0.75 g	Water to 100 ml	
Water to 100 ml: Filter			

* Bis: N,N'-methylene-bis-acrylamide
\+ Tris: Tris(hydroxymethyl)aminomethane
or, 2-amino-2-hydroxymethyl-1,3-propanediol
± Temed: N, N, N', N'-tetramethylethylenediamine

7 mm teflon-coated stirring bar was placed in the flask and the solution stirred until a uniform suspension was achieved.

A drop of 0.1% aqueous methyl green dye was introduced to the top of the gel to act as an acidic tracking dye. Bromophenol blue (0.01%) was used as a basic tracking dye. The protein sample was pipetted into the tube to the required volume. The remainder of the tube was filled with buffer and a 1 mm slice of 10% gel placed on top to prevent loss of sample into the buffer reservoir. Care was taken at all times to avoid bubbles. The paraffin film seal was removed before inserting the tubes into the apparatus.

Electrophoresis was performed in a Canalco disc electrophoresis instrument containing open buffer reservoirs.

The prepared tubes were pushed up into numbered grommets, unused apertures were closed off, and enough buffer added to both reservoirs to cover the ends of the tubes and the electrodes.

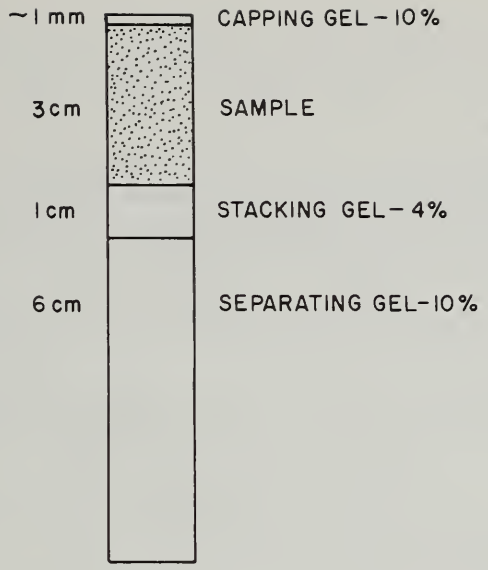

DIAGRAM OF GLASS TUBES USED
FOR SEPARATING SUGAR PROTEINS

FIGURE 1--Diagram of glass tubes used for separating sugar proteins.

Table 3 shows the formulations of the buffers used. Prior to use, 100 ml of the buffer stock solution was diluted to one liter.

The separation was carried out in the refrigerator to avoid damage to the samples due to heat build-up. A constant current of 3 mA per tube was applied, across a potential difference of about 300 V. The pH of the buffer determined the placement of the electrodes, as the protein takes on the sign of the buffer. In an acid electrophoresis, the negative electrode was placed at the bottom reservoir; in a basic separation, the leads were reversed.

TABLE 3. Buffer stock solutions

pH 3.5		pH 8.9	
glycine	14.1 g	glycine	28.8 g
acetic acid	10.8 ml	tris	6.0 g
water to 1000 ml		water to 1000 ml	

Electrophoresis took five hours or longer in an acid buffer system and one to two hours in a basic buffer.

When the separation was completed, the gels were removed from the tubes by reaming the ends of the tubes with a blunt needle attached to a deionized water supply so that a gentle stream of water was emitted to help dislodge the gel from

the tube. The gels were placed at once in 0.05% Coomassie Brilliant Blue R-250 (C.I. 42660) in 12.5% trichloroacetic acid (4) overnight. The next day, the stain was discarded and the gels washed gently once in 50% ethanol to remove insoluble dye that adhered to the sides of the gel and the test tube and to enhance destaining. Removal of absorbed background stain was done in 10% trichloroacetic acid with several solution changes per day for two days. The proteins appeared as distinct blue bands against a clear background when destaining was completed.

Measurement of Protein in the Gels

A colorimetric method based on the elution of the protein-complexed dye from the gels was developed for the determination of the protein in the bands. Each stained band was carefully cut from the gel with a stainless steel razor blade, sectioned into small pieces, and dropped into a 2 ml volumetric flask. About 1 ml of the eluting solution was added (95% DMSO - 5% acetic acid v/v), and the stoppered flasks put on a wrist-action shaker overnight. The next day the solution in the flasks was brought up to volume and the absorption read at 590 nm on a Beckman DB Spectrophotometer. The protein values relative to ribonuclease were taken from the ribonuclease standard curve.

Ribonuclease Standard Curve

Ribonuclease A, molecular weight 13,700, from Iso-Labs was used. The calibration curve was done at pH 3.5 and all subsequent quantitative work was done with this buffer.

Stock solutions containing 1 mg/ml of ribonuclease in diluted buffer were used to make a series of dilutions to obtain the desired quantity of protein per gel, from 1 µg to 100 µg. The procedure outlined earlier for electrophoresis and elution of the dye was used, with the exception that volumes of 5 ml of DMSO-acetic acid as well as 2 ml were used for elution of the dye. The absorbance of the different eluting volumes per weight of protein was related by determining total absorbance units by the following formula:

$$\text{Total absorbance units} = (\text{absorbance of sample at 590 nm}) \times (\text{ml of eluting solvent}).$$

RESULTS AND DISCUSSION

Initially, the proteins were examined in a basic buffer system, but this had the disadvantage that the negatively charged polysaccharides and colorants in the sample also moved into the gel. This problem could be eliminated by electrophoresing the samples in acid buffer with the negative lead in the bottom reservoir. The protein separation remained the same in both buffers, indicating that the proteins had reversed charge from negative to positive in going from basic to acidic buffer. Figure 2 shows the alcohol precipitate of a raw sugar electrophoresed at pH 8.9 and pH 3.5. The two protein bands are characteristic of all raw sugars examined. The faster-moving of the two bands has been designated "protein 1" and the protein with the lesser mobility is called "protein 2." It has also been found that protein 2 disappears during refining and is usually absent in refined sugars. Protein 1 is present in every refined sugar examined,

including those that have undergone additional purification procedures. Figure 3 compares a refined sugar to a raw sugar. It is evident that the single refined sugar band corresponds to protein 1 of the raw sugar.

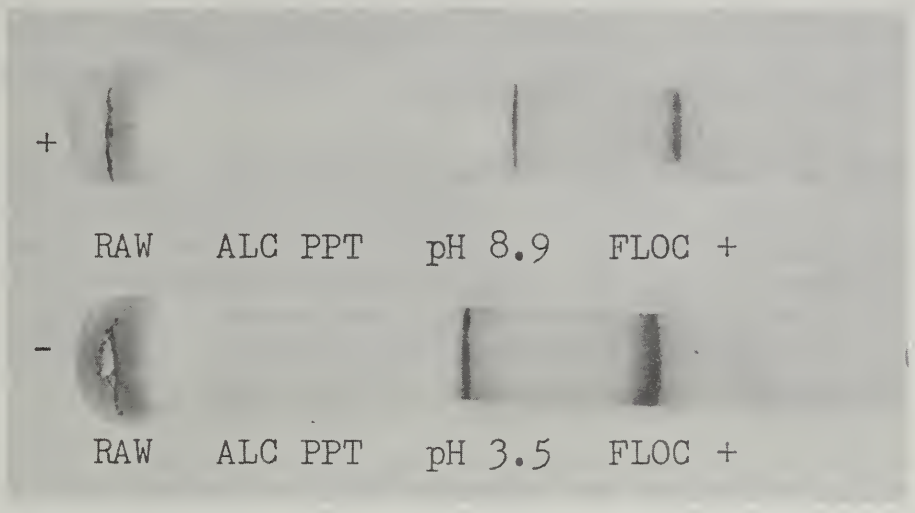

FIGURE 2--Electrophoresis of the alcohol precipitate of a raw sugar electrophoresed at pH 8.9 and 3.5.

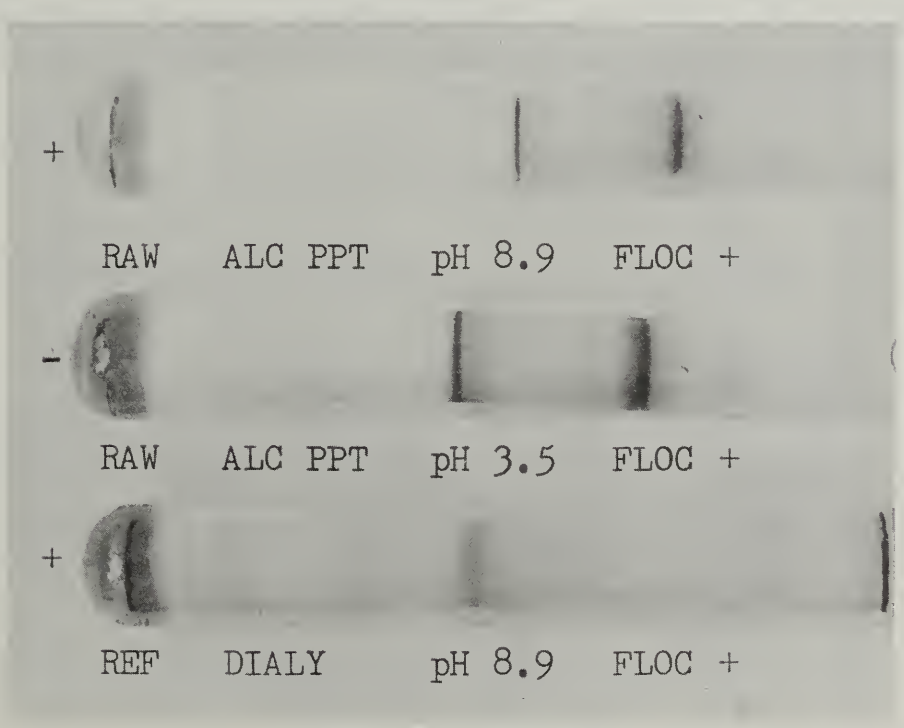

FIGURE 3--Comparison of electrophoresis of the protein in refined sugar and in raw sugar.

Table 4 lists the values found in several sugars. It shows the degree of reproducibility that can be expected with the method.

Table 5 lists the results obtained when a series of refinery samples were analyzed. The results show a steady decrease in protein 1; the results for protein 2 are less predictable. However, protein 2 was removed from the refined sugar as well as from the runoff syrup, and it is assumed that char filtration is responsible for the removal of this protein.

It appears that all stages of refining are efficient to some degree in removing protein 1. The most effective stages are affination and crystallization. Phosphatated liquors were not examined, and so there is no estimation of removal by phosphatation. Affination removed slightly less than half of the protein. The No. 1 runoff syrup contained 4 times as much protein as did the first-strike sugar, indicating the great effectiveness of crystallization in removing this constituent.

Filtration at 80° C through filter aid had little effect. Hot filtration of floc-positive sugar solutions was shown by Roberts, et al. (15) to have no effect on preventing flocculation, whereas cold filtration did prevent floc formation.

The highest protein content was found in raw sugar, particularly in one sample which contained 7.24 µg protein/mg in the nondialyzable fraction. Table 6 shows the amount of protein per band in four separations of this sugar. Table 7 gives the total protein in this sugar based on these separations. The reproducibility was 8.3% with a standard deviation of ±0.6 µg.

Since, with the exception of this unusual raw sugar, none of the bands exceeded 5 µg of protein, the ribonuclease standard curve shown in Figure 4 was used. (A discussion of the limits of linearity of Coomassie Blue in quantification of proteins follows in another section.)

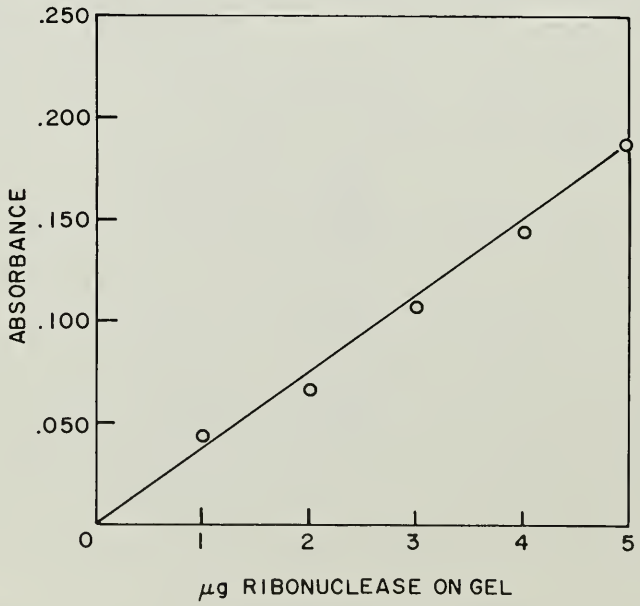

FIGURE 4-- Response of Coomassie Blue binding to ribonuclease.

TABLE 4. Protein in raw and refined sugars measured by gel electrophoresis

Sugar	Protein 1 (ppm)	Protein 2	Total protein on gel
Panama Raw	0.38	0.35	0.73
"	0.41	0.33	0.74
"	0.45	0.29	0.74
Refined Sugar,	0.135	-	0.135
Commercial Blend	0.11	-	0.11
"	0.10	-	0.10
1st Strike Sugar	0.012	-	0.012
"	0.011	-	0.011
"	0.011	-	0.011
4th Strike Sugar	0.036	-	0.036
"	0.028	-	0.028
Refined, Std. Gran.	0.023	-	0.023
Refined, Special Prep.	0.033	-	0.033
Refined Sugar,	0.034	-	0.034
Commercial Blend	0.047	-	0.047

TABLE 5. Protein in refinery samples measured by gel gel electrophoresis (ppm)

Sample	Protein 1	Protein 2
Raw sugar	0.66	0.58
Washed raw sugar	0.38	0.17
Melt liquor mixture	0.19	0.44
Carbonatated liquor, before filteraid filt'n.	0.096	0.052
As above, after filt'n.	0.083	0.13
No. 1 runoff syrup	0.066	-
1st strike sugar	0.016	-

TABLE 6. Protein in the nondialyzable portion of a raw sugar measured by gel electrophoresis

Volume of Eluting Solvent (ml)	µg of protein		Sample size (mg on gel)
	Protein 1	Protein 2	
2	2.75	5.0	0.965
5	2.50	4.0	0.965
2	4.6	8.3	1.93
5	5.0	8.3	1.93

TABLE 7. Concentration of protein in the nondialyzable fraction of a raw sugar

Sample No.	Conc. of protein (µg/mg)
1	7.36
2	6.68
3	8.03
4	6.89

Effect of the Protein on Floc Formation

In his discussion on the role of charged colloidal particles in floc formation, Roberts (14) described an experiment in which the protein isolated from a floc-positive sugar was shown to initiate flocculation in a floc-negative sugar.

In a similar experiment in the present study, it was demonstrated that either protein 1 or 2 will initiate flocculation. Twelve tubes containing protein from the above mentioned raw sugar were electrophoresed, and the two proteins were eluted from the gels by macerating in water and removing the gel by centrifugation. The isolated proteins were put into two solutions containing 0.1 g of indigenous sugarcane polysaccharide (ISP) in 500 ml of distilled water at pH 1.5. Flocculation occurred within two days in both solutions, indicating that both proteins are capable of initiating floc formation. Flocculation did not occur with ISP alone in solution or with added silicate nor did it occur with protein alone in solution or with added silicate.

Some Observations on Protein 2

Protein 2 gave the more diffusely stained band and was the less reproducible of the two proteins on quantification. It is assumed to be the larger protein because of its slower mobility.

Alcian Blue is known to be a polysaccharide stain and has been used to determine the presence of carbohydrate in protein (i.e. , glyco-proteins). Two published methods (3,21) were modified for this study to stain the bands in the above-mentioned raw sugar: protein 2 was positive to Alcian Blue staining by both methods, indicating it is a glycoprotein or, more likely, a protein-polysaccharide complex. Protein 1 was only faintly positive in one of the methods and did not stain at all in the second method.

Another point of interest regarding protein 2 is that when it was placed in 10% trichloroacetic acid overnight, it became opaque and clearly visible to the unaided eye. Protein 1 did not exhibit this phenomenon.

Coomassie Blue as a Quantitative Protein Stain

Coomassie Brilliant Blue R-250, also known as acid blue 83, was found to be particularly suited as a protein stain by Fazekas de St. Groth and coworkers (8). It is a triphenylmethane dye, which in acidic medium is electrostatically attracted to the protonated amino groups (NH_3^+) of the protein. The resulting dye-protein complex is strongly bound, yet fully reversible under appropriate conditions. Its maximum absorption wavelength is 550 nm (8) but was observed to shift in DMSO to 590 nm. It has been demonstrated that pyridine will also cause a shift of the maximum, to 605 nm. (9).

Most of the quantitative work involving Coomassie Blue for measuring proteins in gel electrophoresis has utilized densitometry; the range over which Beer's law is obeyed is limited but has not been definitely established. St. Groth (8) found the linearity of Coomassie Blue to be 0.5 µg/cm to 10 µg/cm

on cellulose acetate for lysozyme and ovalbumin and 0.5 µg/cm to 15 µg/cm for bovine and human serum albumins. Fishbein (10) reported linearity over a range of 1 µg to 50 µg protein in acrylamide gel slabs by densitometry with an accuracy of ±15%. Bickle and Traut (2) reported the range to be 1 µg to 10 µg by densitometry.

The only reference found to the quantification of stained proteins by elution of dye is the work of Fenner, et al. (9), in which they reported using 25% pyridine in water to extract the dye. Their method extended the range of linearity up to 90 µg.

In the present study, ribonuclease concentrations from 1 µg to 100 µg were measured by the dye elution method. The results are shown in Figure 5. Although linearity extended only to 8.0 µg, the points along the curve did show a fair degree of precision and may possibly be used in semiquantitative work. Figure 5 shows the excellent linearity achieved within the required range of 0 µg to 5 µg ribonuclease.

Different proteins will give slightly different curves (4,8,9) with Coomassie Blue because of variations in dye-binding sites, and the exact quantification of proteins cannot be done unless identical reference proteins are available. Therefore, when using a standard protein such as ribonuclease to measure protein concentration, it must be emphasized that the measurements are in terms of "ribonuclease units" rather than absolute protein values.

Coomassie Blue is an outstanding protein dye because of its much higher sensitivity relative to other dyes. Its greatest disadvantage lies in the relatively weak absorptive complex it forms with a protein, which certainly affects reproducibility of any quantitative method.

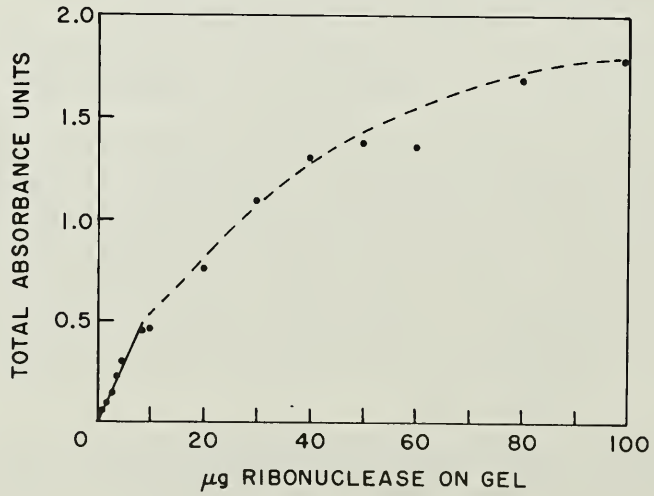

FIGURE 5-- Ribonuclease standard curve.

SUMMARY

The proteins in raw and refined sugars were separated by dialysis, and individual proteins were examined by means of polyacrylamide gel electrophoresis. Semiquantitative measurement of the proteins separated was possible using a colorimetric method based on elution of the protein dye, Coomassie Blue R-250, from the gels.

The quantities of protein found ranged from slightly more than 1 ppm in raw sugars to 0.011 ppm in refined sugar.

The separated proteins were found to cause flocculation in sugar solutions. The efficiency of selected refinery processes in removing the proteins was discussed.

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L. Anhaiser (Imperial): I notice that you were reporting levels of protein down to 0.01 ppm. in refined sugar. Can you tell me how low a level of protein you think you can measure?

M. A. Godshall: We have determined that we can measure as low as 0.002 ppm. This is well below the level that we have found in any sugar.

M. C. Bennett (Tate & Lyle): This has been a very elegant investigation into a very discrete portion of the impurities in sugar. They have been studied from the point of view of the problem; that is, the problem of floc formers, which is of such concern to bottlers. But it is interesting to comment that these impurities, far from being a problem, may in fact be regarded as an essential impurity for some processes which are used in sugar manufacture; for example, juice clarification. We have the belief that the juice clarification process wouldn't work at all if those proteins weren't there. They are probably glycoproteins with fairly low isoelectric points. The same goes for phosphate clarification as practiced in the refinery. I doubt very much whether this process would work as it does if these proteins were not there. The protein seems to be an essential link between the impurity particles and the phosphate flocculation matter which the system has generated. In carbonatation, also, I don't believe the process by which the chalk crystals clump together to form particles which are large enough for us to filter would occur if these proteins were not there. They are an essential part of the flocculation processes which we make use of in our raw and refining manufacturing operations, so it should, perhaps, not be surprising that they do take part in the floc formation which represents a problem to some of our customers.

D. H. Foster (SRI): Why did you choose ribonuclease as the standard?

M. A. Godshall: From its electrophoretic mobility, it was apparent that the sugar protein was small. Ribonuclease is a well-defined, small protein with similar mobility, which fulfilled our requirements for a standard.

N. H. Smith (C&H): Do you have any comments on the relation between molecular size and electrophoretic mobility? In other words, how small does a protein have to be before it moves so fast that it goes all the way through your gel and you don't see it?

M. A. Godshall: Since these samples are separated by a dialysis membrane with a 12,000 molecular weight cutoff, we know there is nothing smaller than 12,000 molecular weight. Ribonuclease A has a molecular weight of 13,700, and the electrophoretic mobility is very rapid, along with the dye front; indicating that any smaller molecules would probably not be retained on the gel. We have separated proteins ranging up to 68,000 molecular weight on these gels, and could probably separate larger molecules as well.

M. Matic (S.M.R.I.): You mentioned that by filtering at low temperature you could eliminate the proteins, but when filtering at high temperature they pass through the filter medium. What is the explanation of that? Are the proteins degraded by heating?

M. A. Godshall: I don't think this small protein is degraded by heating. I think it is associated with a polysaccharide that is colloidal at room temperature and solubilized at a higher temperature.

M. Matic: What one does in the normal juice clarification is to heat above 80°C to precipitate protein. Apparently just the opposite is happening here.

M. A. Godshall: Many proteins are coagulated and removed by heating; however, some plant proteins are heat stable, especially in alkaline and neutral solutions, and these could remain in the process liquors.

M. Fowler (Amstar): How pH sensitive is the blue stain for protein?

M. A. Godshall: Coomassie Blue is a pH sensitive dye and needs to be in the anionic state in slightly acidic media in order to bind to the protonated amine groups of a protein.[1/] This complex can be broken by increasing the pH. The dye is also sensitive to strongly acid or alkaline reagents and is completely decolorized by 2N NaOH, concentrated HCl, 2N H_2SO_4 and phosphonic acid.

J. F. Dowling (Refined Syrups): When you put the protein through the Millipore filter of 0.22 or 0.45 mμ, did you put it through with the polysaccharide or did you put the protein through by itself?

M. A. Godshall: The entire sugar solution was filtered.

J. F. Dowling: So what was held back could have been the combination of the protein and polysaccharide and not just the protein. The protein and polysaccharide could have been complexed and the combination of the two made a big enough molecule to be held on the filter. Upon heating, the polysaccharide will go through; in a cool solution the polysaccharide is held. If the protein and polysaccharide are combined, they are held when cool and can be eluted with hot water. I doubt that the material on the filter is the protein by itself.

M. A. Godshall: We feel that the protein is complexed with the polysaccharide.

M. Matic: Does this mean that the protein polysaccharide link is broken by heating?

E. J. Roberts (CSRRP): The dynamic energy put into the protein-polysaccharide complex by heat could overcome the electrostatic attraction of the two. Then they might pull apart and consequently go through the filter. That's a speculation.

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THE ROLE OF CHARGED PARTICLES IN FLOC FORMATION

By Earl J. Roberts and Mary An Godshall[1]

ABSTRACT

Further studies on the floc-causing factors in cane sugar have revealed that these substances are colloidal in nature. The electrostatic charges on these substances (polysaccharides and proteins) have been studied over a wide pH range. It was found that the negatively charged polysaccharides and the positively charged protein at low pH are key factors in floc formation.

INTRODUCTION

Acid beverage floc, a white feathery precipitate, that is occasionally found in carbonated beverages and acidified sugar sirups, has been a source of trouble for sugar refiners for many years. Although many refiners have conducted research on the cause of floc formation, little of this work has been published. Results, including those that have been published, are contradictory and inconclusive.

Stansbury and Hoffpauir (10)[2] found that sugarcane floc consisted of starch, lipids, protein, silicates and decolorizing carbon. They reported a high correlation between the quantity of carbon and the amount of floc formed. Cohen et al. (1) reported that acid floc contained protein, inorganic material and a starch-like polysaccharide. They concluded that the polysaccharide bore no special relation to floc formation but they reported a high correlation between the amount of protein present and the amount of floc formed.

Roberts and Carpenter (7) reported that isolated floc was composed of protein, silicon dioxide, lipids, starch and a polysaccharide composed of arabinose, rhamnose, mannose, galactose and glucose. Miki et al. (6) reported the presence of an identical polysaccharide in isolated beverage floc.

Liuzzo and Huan-Wen Hsu (5) studied the alcohol floc from cane sugar and concluded that the floc is caused primarily by an amylose-related substance that can complex with other compounds to enhance floc formation, and that protein and peptides are among the complexing materials that contribute to the formation of the aggregate.

This paper describes what the authors believe to be the cause of floc formation in acidified cane sugar solutions along with methods for removal of the causative substances.

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[2]Numbers in parentheses refer to items under "References" at the end of this paper.

EXPERIMENTAL

Sugars used were raw sugars from various sources, fourth, third, second, and first strike sugars, and commercial blends. The tests used to determine the floccing characteristics of sugars were the Coca Cola floc test (3) and the beverage test (4). In both tests an acid solution of the sugar is examined visually for many days for the onset of floc.

The indigenous sugarcane polysaccharide (ISP) was prepared as described by Roberts et al. (9) from fresh sugarcane by a method that excludes starch and dextran.

Removal of Floc Causing Material from Sugar Solutions

Several solutions were prepared by dissolving 500 grams of floc-positive sugar in 500 ml water. Each solution was filtered at 25°C through a 10 mm mat of Celite analytical filter aid at pH values ranging in steps of 1 pH unit from 1.5 to 8.5. The pH was then adjusted to 1.5 to 2 with phosphoric acid. If, after ten days no floc had formed, it was assumed that the floc-causing material had been removed by the tight filtration.

A solution of 500 g of floc-positive sugar in 500 ml of water was centrifuged at 40,000 x G. The pH of the effluent was adjusted to 1.5 to 2.0 with phosphoric acid. If, after ten days no floc had formed, it was assumed that the floc-causing material had been removed.

Nature of the Charge on Sugarcane Polysaccharides

Twenty-five ml of 0.1 M phosphate buffer was placed in a U-tube. A solution of 0.5 g of ISP in 25 ml of the same buffer was carefully added through the bottom of the U-tube so that the boundary between the turbid ISP solution and the nonturbid buffer was clearly visible in both arms of the U-tube. An electrode was placed in each arm of the U-tube, a potential of 200 volts DC was applied across the tube, and the boundary between the ISP solution and the buffer was observed. The experiment was repeated with buffers of several pH values ranging as low as pH 3.0. In each case, ISP solution moved quickly toward the positive electrode, indicating a strong negative charge on the ISP.

Precipitation of Polysaccharide by Positively Charged Compounds

To 100 g of floc-positive sugar dissolved in 100 ml water, without pH adjustment, was added 10 ml of saturated solution of Basacryl Orange FL dye (strong positively charged basic dye). Floc formed in six hours. It was also found that the addition of 10 ml of dye solution to a solution of 0.2 g of ISP in 100 ml of water caused immediate precipitation of polysaccharide.

Isolation of Protein from Sugar

By dialysis

Two hundred grams of the sugar to be analyzed were dissolved in 500 ml of distilled water and placed in a tube of regenerated cellulose with a cut-off of 10,000 to 12,000 molecular weight. The cellulose tube containing the sugar

solution was placed in a glass tube on a rocking dialyzer and dialyzed against flowing toluene-saturated water for 100 hours. After dialysis, the solution remaining in the bag was concentrated at a temperature below 60 degrees in the rotary evaporator, and then freeze dried.

By ion exchange

A solution of 500 g of floc positive sugar was dissolved in 1500 ml of deionized water. The solution was percolated through a 20 mm i.d. column containing 100 g of Dowex-2 anion exchange resin in the base form. The resin was washed with 100 ml of deionized water to remove sugar, and then washed with 500 ml of 10% sodium chloride solution to recover the protein. The sodium chloride was removed by dialysis for 72 hours against toluene-saturated deionized water.

Floc Formation with Isolated Protein and ISP

The protein isolated from 500 g of floc positive sugar by ion exchange was dissolved in 2000 ml of water. To one half of this solution was added 0.2 g of ISP and to the other half of the solution was added 0.2 g of ISP and 100 g of floc negative sugar. The pH of each solution was adjusted to 2.0 with phosphoric acid. Floc formed in both solutions within two days indicating that the combination of the protein and ISP certainly caused floc.

Quantitative Determination of Sugar Protein

The proteins which were separated from the sugar by dialysis were determined as described by Godshall and Roberts (4).

RESULTS AND DISCUSSION

Removal of Floc-Causing Materials from Sugar

The fact that the floc forming materials can be removed by filtration and centrifugation show that these substances are colloidal in nature rather than in true solution. Their presence accounts for at least part of the turbidity in sugar solutions. Analysis of the materials removed by these methods shows that their composition is similar to that of isolated floc.

One of the major components common to all of these products, including isolated floc, is a heterogeneous polysaccharide that is indigenous to sugar cane (ISP). In a recent publication the composition of this polysaccharide was described (8).

Nature of the Charge on ISP

It has been known for many years that the colloids in cane juice carry a negative charge at normal juice pH. Since it had been shown that both the isolated floc and the floc-causing material removed from floc-positive sugar by centrifugation or filtration contained ISP, it was of interest to determine the charge on isolated ISP.

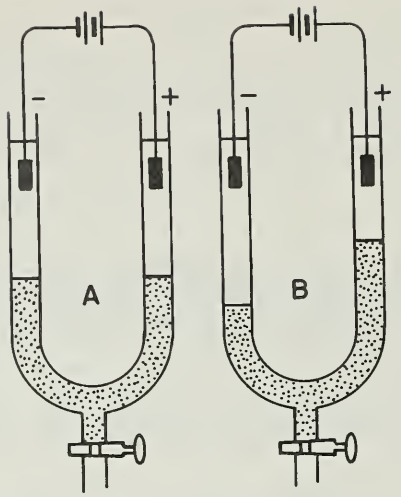

FIGURE 1--A diagrammatic representation of electrophoresis
A--Boundaries before electrophoresis
B--Boundaries after electrophoresis.

ISP in solution was observed to migrate toward the positive electrode when a potential difference was applied across the tube containing the solution. Figure 1 shows the position of the boundaries between the ISP solution and the buffer before and after electrophoresis. The direction of the boundary change indicates that the ISP carries a strong negative charge over a wide pH range. This conclusion was substantiated by the fact that ISP was precipitated by compounds carrying a strong positive charge, such as Basacryl Orange FL dye.

It has long been known that flocculation occurs when a negatively charged colloid is mixed with a positively charged one. Flocculation was found to be nearly complete in those cases where the numbers of opposite charges were nearly the same. Such reactions have been observed with a variety of systems, and may be illustrated as follows:

$$(\text{Colloid})_a^+ + (\text{Colloid})_b^- \longrightarrow \underline{(\text{Colloid})_a(\text{Colloid})_b} \downarrow$$

Cane Sugar Protein

The fact that ISP is precipitated by compounds carrying a positive charge suggests that floc-positive sugars may contain some substance capable of changing its charge from negative in alkaline and neutral solution to positive in acid solution. It is well known that the charge on many proteins in solution changes from negative in alkaline and neutral solutions to positive in acid solutions. Godshall and Roberts (4) have shown that protein isolated from cane sugar changes its charge from negative at pH 8.9 to positive at pH 3.5. The addition of cane sugar protein to a solution of ISP at pH 1.5 to pH 2.0 produced floc both in water and in sugar solution. This confirms the finding that the charge on the sugarcane protein changes from negative in alkaline and neutral solution to positive in acid solution and that the combination of the two colloids of opposite charges causes flocculation. Based upon the foregoing experimental results, it seems reasonable to conclude that the basic cause of floc

formation in acidified cane sugar solutions is the combination of ISP and protein. Although several other components present in sugars, such as silicon compounds, starch, dextran and lipids, are negatively charged, the charges are weak and these compounds alone or in combination with ISP do not cause floc, as far as can be determined. It is postulated that floc formation is preceded by aggregation of oppositely charged colloids, the aggregates then coalesce to form the floc particle. As this process occurs, many of the other colloidal materials present are adsorbed onto the floc particles and are carried along with it. The nature of the colloids in the sugar may determine the appearance of the floc: for example, a high starch sugar may form "cotton ball" floc while a high silicon sugar may form a "granular" floc.

Many samples of sugar do not form floc when their solutions are acidified even though they contain both ISP and protein. In sugars of high purity the concentration of ISP and/or protein may be so low that too few collisions occur between oppositely charged particles to cause aggregation.

Deuel et al. (2) have shown that maximum flocculation occurs when the numbers of positive and negative charges are nearly equal. In solutions in which there is a preponderance of negatively charged particles the positively charged particles may become surrounded by negatively charged ones so that the overall charge on the complex particle is negative. This phenomenon is illustrated in figure 2. In such cases flocculation would not occur. This type of condition may explain why some low purity sugars do not form floc. In some cases the electrostatic characteristics or structure of the ISP or protein may be altered during the manufacture and refining of sugar, and thus the conditions necessary for floc formation may be destroyed.

SUMMARY

It has been shown that all cane sugars contain a heterogeneous polysaccharide which is indigenous to sugarcane. This polysaccharide forms a colloidal solution which carries a negative charge over a wide pH range. It was also shown that refined cane sugar contains protein which changes its charge from negative in alkaline and neutral solutions to positive in acid solutions. Combining solutions of the isolated polysaccharide and the isolated protein at low pH in the presence or absence of sugar results in floc formation. It seems

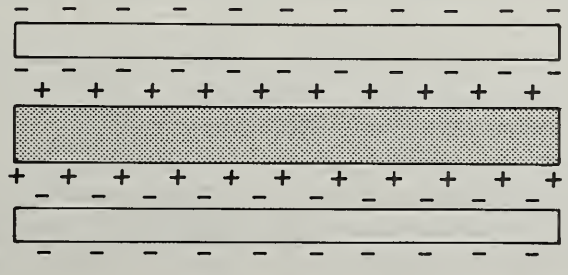

FIGURE 2--A complex particle composed of one positively charged component and two negatively charged components.

reasonable therefore to conclude that the indigenous polysaccharide and protein in cane sugar are the basic factors that cause floc formation in acidified sugar solutions.

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DISCUSSION

R. L. Knecht (Colonial Sugars): Your previous papers on surveys of actual inplant removal of polysaccharide did not show much polysaccharide removal by ion exchange. Can you comment on that? Are the ion exchange columns quickly overloaded?

E. J. Roberts: Probably the ion exchange column is soon overloaded. The hexuronic acids in the polysaccharide are very weak acids, that is compared to phosphoric acid; so I think the columns are easily overloaded. Perhaps, in our

survey paper, the resin was a weak anion resin that would also account for low removal.

S. E. Bichsel (American Crystal): The ISP seems to contain saccharate units and possibly hexuronic acids. Would you care to comment on whether or not this is pectin?

E. J. Roberts: As all probably know, it has been reported since the year 1 that sugarcane contains pectin. But, I've never seen any evidence of it yet. If you hydrolyze sugarcane polysaccharide with acid, you won't find any galacturonic acid, which is one of the necessary components and hydrolysis products of pectin. The reason is that the glycosidic linkage in galacturonic acid is so strong that it is not hydrolyzed under ordinary conditions. If you use conditions stringent enough to hydrolyze the glycosidic linkages in the galacturonic acid you also decarboxylate it. So you still don't see galacturonic acid. When we treated pectin with pectinase, we formed galacturonic acid. When we treat the polysaccharide with pectinase we don't find any galacturonic acid. So, my assumption is, that if there is no galacturonic acid, the polysaccharide is not pectin even though it is a pectin-like substance.

W. C. Strunk (FMC Corporation): It would appear that the amount of protein needed to bring down this floc is pretty small compared with the amount of the polysaccharide present. Do you think that this is a function of charge density or of molecular weight, or what do you think is the reason for this?

E. J. Roberts: I think it is a function of charge density. Proteins are composed of individual amino acids, and for each amino acid you find a certain charge. The hexuronic acid is responsible for the charge on the polysaccharide and apparently there are only a few hexuronic acid units per molecule. The purest sample of polysaccharide that we could prepare contained only 8.5% of hexuronic acid. The non-dialyzable portions of most sugars contain about 3.5% hexuronic acid. This indicates that the ISP in the non-dialyzable fraction is diluted with starch, dextran, silica, and whatever other large molecules are in there. So there are probably many more charges per unit weight on the protein than on the polysaccharide.

J. F. Dowling (Refined Syrups): From how many origins were the raw sugars that you looked at in the last four years that were floc positive?

E. J. Roberts: About three or four different origins gave floc positive sugars.

J. F. Dowling: We must get more samples from everyone when there is a floc problem. The thing that we get into here is that if you take sugars from only one country of origin, or region then all that you have identified is the floc from that one place. When anyone here has a floc problem, I think that it is very important that he send a sample to Earl so that we can get the whole picture. The samples that C.S.R.R.P. has been working on have been limited basically to a few countries, and we want to spread this out.

As I see it right now, economically, we cannot remove floc with ion exchange resins. The amount of floc there is so small that we would pay a high price to remove it to 0.1 ppm. So, the only answer that I see now is that the

person who grows the cane has to grow it under the right conditions, so that the polysaccharide, per se, does not form in a concentration that we cannot take out in the present process. We must go back to the origins that are growing the floccing sugar. The growers will have to alter the growing conditions or whatever it takes to solve the problem for refiners.

E. J. Roberts: In working with floc you have to rearrange your thinking in terms of quantities of fractions of ppm, because that's the level of these things.

M. Matic (SMRI): I would like to congratulate you and your coworkers on a marvelous piece of work - I think this is a great breakthrough which may possibly indicate the way to eliminate floc. In this connection, for removing your floc, you used ion exchange. I suppose this was done at room temperature, but every refiner runs ion exchange at about 80°; would everything go through at this temperature?

E. J. Roberts: We have not tried it above room temperature.

M. Matic: I believe that what you have done so far is to put two, in my opinion, most important points together. To induce coacervation, you need two things: one is protein which is going to change its charge at certain pHs, and the other thing is a polysaccharide having acidic groups. In view of this, the hexuronic acid component may be the clue to floc formation, and possibly is the reason why other polysaccharides do not form floc because they are lacking acid groups to react with the protein.

M. C. Bennett (Tate & Lyle): A thought crossed my mind while listening to your presentation. The problem that the bottlers experience is not so much that these proteins and polysaccharides are present but that they actually coagulate and visually appear. It really doesn't matter whether they are there or not. What matters is that they appear. Reference was made to some basic principles of colloid stabilization and destabilization, and you rightly pointed out that you could just as well protect the colloid as destabilize and precipitate it. That, therefore, brought to mind the possibility that instead of looking for methods of removing either one or the other, or both, can you not look for something which actually protects the colloid from precipitating, something you can actually put in to stabilize the whole system. You are already talking about ppb levels, so therefore one would only be considering ppb of additive. Just off the cuff, one could think of a whole range of materials, for example highly solvated polysaccharides, polyphosphates and things like that. Do you think that idea is worth exploring?

E. J. Roberts: We had thought of that idea, and will try it. It was only a few days ago that we confirmed that this thing had acid groups on it. We had for a long time assumed that it didn't since it didn't respond to pectinase. I think your idea is a good one.

K. H. Schoenrock (Amalgamated Sugar): We do something like that in the beet sugar industry. We prevent floc formation by boiling at relatively high pH. Of course, we are dealing with a different breed of cat, so to speak. In the beet industry, by boiling at a pH above 8.5, the floc-forming characteristic of the refined sugar is greatly reduced because the saponins stay in solution and go out in molasses.

E. J. Roberts: High pH operation is a problem in the cane industry because of the formation of colorants from the invert sugars.

K. H. Schoenrock: We don't like the high pH either.

J. C. P. Chen (Southdown): In the cane sugar factory we do not usually use a high pH except in carbonatation where the pH may go above 8.5. Would that be the reason why carbonatation removes some of the protein.

E. J. Roberts: I think the *in situ* formation of the finely divided calcium carbonate is responsible for the removal of protein rather than the high pH.

M. C. Bennett: I don't know whether the high pH causes destruction or creation of something.

To keep the floc from forming, I think that I would just go along the shelf and pick some likely chemicals and hope that maybe something would show some sign of inhibiting the floc formation.

F. G. Carpenter (C.S.R.R.P.): We should take a lesson from what we have observed in the laboratory. There are some sugars which are very high in dextran, but do not flocculate. Maybe the dextran is the protecting component which prevents flocculating.

S. E. Bichsel (American Crystal): In the sugar beet purification process we too have a lime and carbon dioxide purification, but, the floc that we deal with may be different from what you're talking about. The beet process is very efficient in floc removal; we eliminate somewhere between 98% and 99% of floc. Carbonatation purification reduces floc down to several hundred ppm in thick juice. This is what we have in our white pan feed on our first strike, but even this is enough to cause a problem. If we do an inefficient job with lime and carbon dioxide purification, or run a cloudy clarifier, then we know that the increased amount of floc going to the first strike will definitely cause problems.

K. C. Leverington (Bureau of Sugar Experiment Stations, Queensland): Our plant breeders would say that if you were to throw another criterion on the plant breeding selection, that would mean that we would get no new varieties for 100 years.

MICROBIOLOGY OF SUGAR: A TAXONOMIC STUDY

By Richard D. Skole, J. N. Hogu, and Anthony B. Rizzuto[1]

(Presented by R. D. Skole)

ABSTRACT

This paper will present a general review of microorganisms common to the cane sugar refining process and discuss the practical application of these findings to a study involving the enumeration of aerobic mesophilic microorganisms in sugar products.

INTRODUCTION

During the past fifteen years, the Microbiology Section at Research and Development, Amstar Corporation, has accumulated extensive taxonomic data on bacteria, yeasts, and molds isolated in the refining process, raw sugars and finished sucrose products. The term "taxonomy" may be defined as the classification of plants and animals according to their natural relationships.

Specific identification of microorganisms is a difficult task involving the consideration of many factors. Included in these factors are the observations on the microscopic as well as macroscopic characteristics of the organism. Thus the size, shape, color, and texture of the colony when grown upon various different nutrient substrates must be considered as well as microscopic determinations of the size, shape, internal and external physical structures of the individual cells comprising these colonies. Finally, specialized determinations upon the biochemical and physiological capabilities of the organism must be made in order to classify the organism with any degree of assurance.

Basically all microorganisms are classified according to this general procedure although investigators concerned with identification of the different groups (bacteria, yeasts, and molds) consider certain observations to be more important than others. Thus in identifying bacteria and yeasts, more weight is placed upon microscopic and physiological determinations than colonial morpho - ogy, while molds generally are classified on the basis of gross colonial characteristics.

The primary objective of this taxonomic program was to establish a common microflora in sugar refining. This has an important practical consideration as the so called sucrose-organism complex is a reality that every refiner by necessity must fully appreciate, if maximum microbial control is to be maintained on a daily basis. Identification of the microflora in sugar refining will help immeasurably to guard products against premature spoilage, to reduce sucrose loss caused by biological action, and to increase the general sanitation of the working environment.

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The first studies of microorganisms associated with sugar refining were conducted over a hundred years ago (3,5,6,)[2]. It is beyond the scope of this paper to review all past work on the various microorganisms identified in raw sugar, in the refining process or sucrose products. Therefore, the following discussion will be restricted to taxonomic studies made in this laboratory of bacteria, yeasts and molds common to sugar refining.

YEASTS

The fungi known as yeasts are organisms of very great economic importance. Certain species are used for the process of baking and the production of alcoholic beverages by fermentation. Others are responsible for the development of special flavors in certain wines after the main fermentation is completed. From their natural habitats yeasts are often carried into the fermentation and food industries, where they may act as spoilage organisms. Often a special flora is found, as with the lactose-fermenting yeasts in dairy products and osmophilic types in substrates with high concentrations of dissolved solids.

The yeasts are spherical, ovoid, or rod shaped ascomycetous fungi, in which the usual and dominant growth form is unicellular. Yeast multiply by budding, transverse fission, or asexual spore formation.

The taxonomy of the yeasts is difficult and only a brief outline on the subject can be given here. The most authoritative monography have come from the Dutch school, working mainly at the Centraalbureau voor Schimmelcultures, Yeast Division, Delft, Netherlands. Recently a new work by J. Lodder (6) discusses in detail the various morphological and physiological properties utilized in the differentiation of yeasts into genera and species. This text was used in all of the present yeast identification studies.

A large number of yeasts (450 cultures) had been isolated in pure culture from various raw sugars and in-process refinery materials for taxonomic studies. The selection of colonies for identification was on a random basis since it was intended to provide a picture of the range of species present rather than quantitative relative distribution. However, since yeasts are one of the few microorganisms with the metabolic capabilities to grow in high concentrations of dissolved solids, our taxonomic studies of these organisms were generally restricted to osmotolerant or osmophilic strains.

Saccharomyces Species

Of the 30 species found in the genus Saccharomyces, one of the most important is Saccharomyces cerevisiae. Strains of this yeast are employed in many food industries, for the leavening of bread, as top yeasts for ale, for wines, and for the production of alcohol, glycerol and invertase.

Also found in the genus Saccharomyces are the majority of osmophilic yeast strains. These yeasts are now classified in only two species and one variety, Saccharomyces rouxii, Saccharomyces mellis, and Saccharomyces var. polymorphus.

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[2]Numbers in parentheses refer to items under "References" at the end of this paper.

TABLE 1--The classification of yeast isolated from various raw sugars

Sample number	Source of culture	Genus and species designation
101-A	Guatemala raw sugar	Saccharomyces cerevisiae
102-A	" " "	Saccharomyces cerevisiae
103-A	" " "	Saccharomyces rouxii
104-A	Philippine Island raw sugar	Saccharomyces cerevisiae
105-A	" " " "	" "
106-A	" " " "	" "
107-A	" " " "	" "
108-A	" " " "	Saccharomyces rouxii
109-A	" " " "	Saccharomyces cerevisiae
110-A	" " " "	" "
111-A	Puerto Rico raw sugar	Saccharomyces mellis
112-A	" " " "	" "

TABLE 2--The classification of yeast isolated from various raw sugars

Sample number	Source of culture	Genus and species designation
101-B	Peru raw sugar	Saccharomyces cerevisiae
102-B	" " "	" "
103-B	" " "	" "
104-B	Dominican Rep. raw sugar	Saccharomyces cerevisiae
105-B	" " " "	" "
106-B	" " " "	Saccharomyces cerevisiae-ellipsoideus
107-B	" " " "	Saccharomyces carlsbergensis
108-B	" " " "	Saccharomyces cerevisiae
109-B	" " " "	" "
110-B	" " " "	Saccharomyces carlsbergensis
111-B	Hawaiian raw sugar	Saccharomyces cerevisiae-ellipsoideus
112-B	" " "	Saccharomyces cerevisiae-ellipsoideus

TABLE 3--The classification of yeast isolated from various raw sugars

Sample number	Source of culture	Genus and species designation
101-C	Hawaiian raw sugar	Saccharomyces carlsbergensis
102-C	" " "	Saccharomyces cerevisiae
103-C	" " "	" "
104-C	" " "	" "
105-C	" " "	" "
106-C	" " "	" "
107-C	French West Indies raw sugar	Saccharomyces cerevisiae
108-C	Mexican raw sugar	Saccharomyces cerevisiae-ellipsoideus
109-C	" " "	" "
110-C	" " "	" "
111-C	British West Indies raw sugar	Saccharomyces cerevisiae
112-C	" " " " "	Saccharomyces rouxii

It is necessary to recognize that definitions vary and that for the most part distinctions between groups of yeasts are purely arbitrary. For the purposes of our taxonomic studies, however, the definition of Scarr (8) for osmophilic yeasts was used. Scarr defined osmophilic yeasts as those which can grow at concentrations over 65 Brix, which state in pure sucrose solutions is near saturation.

The data recorded in tables 1 to 3 shows the several Saccharomyces species identified from various raw sugars. The predominant strain is Saccharomyces cerevisiae followed by the osmophilic strains. Typically the osmophilic strains are in the minority under natural conditions, until by either drying out or some manufacturing process, the water content is decreased, allowing them to multiply.

Of the biochemical (physiological) tests used in the taxonomy of yeasts, carbohydrate fermentation and assimilation of carbon compounds and nitrates are the most important.

Fermentation of carbohydrates by yeasts results in the production of alcohol and carbon dioxide. For the fermentation of a particular carbohydrate, a corresponding enzyme system is required. This is controlled by various genes, and both biochemical and genetic studies have been made on such enzyme systems.

The ability to ferment a given carbohydrate has proved to be rather constant in yeast strains. The fermentation pattern is one of the important characteristics of a specie. For the differentiation of genera the fermentation rate is generally less important, but is often helpful. The carbohydrates generally used in the fermentation studies are glucose, galactose, sucrose, maltose, lactose, and raffinose.

TABLE 4--Gas production recorded in percent by the gasometer scale in carbohydrate fermentation tubes after seven days' incubation at 28°C.

Culture number*	Carbohydrate source						
	Glucose	Fructose	Sucrose	Galactose	Maltose	Lactose	Raffinose
101-A-S.c	100	100	100	95	85	0	5
102-A-S.c	100	100	90	100	55	0	25
103-A-S.r	100	100	0	0	75	0	0
104-A-S.c	100	100	100	90	100	0	35
105-A-S.c	100	100	100	100	100	0	5
106-A-S.c	100	100	90	85	100	0	15
107-A-S.c	100	100	100	95	100	0	30
108-A-S.r	100	100	0	0	50	0	0
109-A-S.c	100	100	100	45	100	0	5
110-A-S.c	100	100	50	100	100	0	20
111-A-S.m	100	100	0	0	0	0	0
112-A-S.m	100	100	0	0	0	0	0

*S.c designates Saccharomyces cerevisiae
S.r designates Saccharomyces rouxii
S.m designates Saccharomyces mellis
Source of cultures refer to tables 1 through 3

TABLE 5--Gas production recorded in percent by the gasometer scale in carbohydrate fermentation tubes after seven days' incubation at 28°C.

Culture number*	Carbohydrate source						
	Glucose	Fructose	Sucrose	Galactose	Maltose	Lactose	Raffinose
101-B-S.c	100	100	100	50	100	0	15
102-B-S.c	100	100	100	100	55	0	0
103-B-S.c	100	100	100	75	100	0	15
104-B-S.c	100	100	100	100	75	0	30
105-B-S.c	100	100	75	100	100	0	5
106-B-S.c.e	100	100	100	100	100	0	10
107-B-S.ca	100	100	100	60	90	0	100
108-B-S.c	100	100	90	100	100	0	5
109-B-S.c	100	100	100	75	100	0	10
110-B-S.ca	100	100	95	100	100	0	100
111-B-S.c.e	100	100	100	100	90	0	25
112-B-S.c.e	100	100	100	100	100	0	15

*S.c designates Saccharomyces cerevisiae
S.c.e designates Saccharomyces cerevisiae-ellipsoideus
S.ca designates Saccharomyces carlsbergensis
Source of cultures refer to tables 1 through 3

TABLE 6--Gas production recorded in percent by the gasometer scale in carbohydrate fermentation tubes after seven days' incubation at 28°C.

Culture number*	Carbohydrate source						
	Glucose	Fructose	Sucrose	Galactose	Maltose	Lactose	Raffinose
101-C-S.ca	100	75	100	85	100	0	100
102-C-S.c	100	100	100	100	85	0	10
103-C-S.c	100	100	75	100	100	0	5
104-C-S.c	100	100	100	90	90	0	30
105-C-S.c	100	80	100	100	100	0	15
106-C-S.c	100	100	90	55	100	0	10
107-C-S.c	100	100	100	100	100	0	0
108-C-S.c.e	100	90	100	60	90	0	0
109-C-S.c.e	75	100	75	100	65	0	15
110-C-S.c.e	100	90	100	75	100	0	30
111-C-S.c	100	100	100	45	35	0	5
112-C-S.r	100	100	0	0	75	0	0

*S.c designates Saccharomyces cerevisiae
S.c.e designates Saccharomyces cerevisiae-ellipsoideus
S.ca designates Saccharomyces carlsbergensis
S.r designates Saccharomyces rouxii
Source of cultures refer to tables 1 through 3

TABLE 7--Sole carbohydrate source in base medium, growth recorded as negative or positive after seven days' incubation at 28°C.

Culture number*	Carbohydrate source						
	Glucose	Fructose	Sucrose	Galactose	Maltose	Lactose	Raffinose
101-A-S.c	+	+	+	+	+	-	+
102-A-S.c	+	+	+	+	+	-	+
103-A-S.r	+	+	-	-	+	-	-
104-A-S.c	+	+	+	+	+	-	+
105-A-S.c	+	+	+	+	+	-	-
106-A-S.c	+	+	+	+	+	-	+
107-A-S.c	+	+	+	+	+	-	+
108-A.S.r	+	+	-	-	+	-	-
109-A-S.c	+	+	+	+	+	-	-
110-A-S.c	+	+	+	+	+	-	+
111-A-S.m	+	+	-	-	-	-	-
112-A-S.m	+	+	-	-	-	-	-

*S.c designates Saccharomyces cerevisiae
S.r designates Saccharomyces rouxii
S.m designates Saccharomyces mellis
Source of cultures refer to tables 1 through 3

TABLE 8--Sole carbohydrate source in base medium, growth recorded as negative or positive after seven days' incubation at 28°C.

Culture number*	Carbohydrate source						
	Glucose	Fructose	Sucrose	Galactose	Maltose	Lactose	Raffinose
101-B-S.c	+	+	+	+	+	-	+
102-B-A.c	+	+	+	+	+	-	-
103-B-S.c	+	+	+	+	+	-	+
104-B-S.c	+	+	+	+	+	-	+
105-B-S.c	+	+	+	+	-	-	-
106-B-S.c.e	+	+	+	+	+	-	+
107-B-S.ca	+	+	+	+	+	-	+
108-B-S.c	+	+	+	+	+	-	-
109-B-S.c	+	+	+	+	+	-	+
110-B-S.ca	+	+	+	+	+	-	+
110-B-S.c.e	+	+	+	+	+	-	+
112-B-S.c.e	+	+	+	+	-	-	-

*S.c designates Saccharomyces cerevisiae
S.c.e designates Saccharomyces cerevisiae-ellipsoideus
S.ca designates Saccharomyces carlsbergensis
Source of cultures refer to tables 1 through 3

TABLE 9--Sole carbohydrate source in base medium, growth recorded as negative or positive after seven days' incubation at 28°C.

Culture number*	Carbohydrate source						
	Glucose	Fructose	Sucrose	Galactose	Maltose	Lactose	Raffinose
101-C-S.ca	+	+	+	+	+	-	+
102-C-S.c	+	+	+	+	+	-	+
103-C-S.c	+	+	+	+	+	-	-
104-C-S.c	+	+	+	+	+	-	+
105-C-S.c	+	+	+	+	+	-	+
106-C-S.c	+	+	+	+	+	-	+
107-C-S.c	+	+	+	+	-	-	-
108-C-S.c.e	+	+	+	+	+	-	-
109-C-S.c.e	+	+	+	+	+	-	+
110-C-S.c.e	+	+	+	+	+	-	+
111-C-S.c	+	+	+	+	+	-	-
112-C-S.r	+	+	-	-	+	-	-

*S.c designates Saccharomyces cerevisiae
S.c.e designates Saccharomyces cerevisiae-ellipsoideus
S.ca designates Saccharomyces carlsbergensis
S.r designates Saccharomyces rouxii
Source of cultures refer to tables 1 through 3

The yeasts identified in tables 1 to 3 as Saccharomyces cerevisiae, Saccharomyces cerevisiae var. ellipsoideus and Saccharomyces carlsbergensis showed wide fermentative capabilities of these yeast species, as shown in data recorded in table 4 to 6. All the carbohydrates undergoing the fermentation tests, with the exception of lactose, showed varying amounts of gas as recorded by the gasometer scale. The yeast species identified as Saccharomyces rouxii fermented only glucose, fructose, and maltose, while Saccharomyces mellis fermented only glucose and fructose.

Carbohydrate assimilation is closely related to the fermentation of carbohydrates and is useful to differentiate species. Assimilation may be defined as the process by which carbohydrates are utilized by yeasts for the building of cellular constituents.

The assimilation of carbohydrates by a yeast can be detected by observing growth in a complete medium containing a particular carbohydrate as sole source of carbon. Both fermentation and respiration may be responsible for this growth.

The data recorded in tables 7 to 9 show how closely the assimilation of a carbohydrate is related to the fermentation patterns of yeasts. With the exception of several negative reactions of maltose and raffinose, the species identified as Saccharomyces cerevisiae, Saccharomyces cerevisiae var. ellipsoideus, and Saccharomyces carlsbergensis show the broad assimilation patterns of these organisms. These species, however, as noted in the fermentation studies, could not assimilate the carbohydrate lactose.

The species identified as Saccharomyces rouxii and Saccharomyces mellis have very limited assimilation capabilities.

Hansenula Species

Hansenula yeasts differ from most other yeasts in their ability to use nitrate as a source of nitrogen and in the shape of their ascospores, usually hat or "saturn" shaped. Wickerham (15) made an extensive study of this genus including several new species which had in common not only the ability to assimilate nitrate, but other properties, such as the formation of a pellicle, pseudo- and true hyphae and no fermentation to vigorous fermentations. The Hansenula genus includes nine species with one variety; the type culture of the genus is Hansenula anomala.

Species of Hansenula yeast are commonly found in the soil, as serious deteriogens in industrial fermentation processes, in exudates and gums of trees and on fresh, dried, or sugared fruits. However, very little has been reported in the literature concerning the occurrence of this yeast genus in raw sugar. Our earlier taxonomic studies of yeast found in raw sugar rarely isolated Hansenula species. In 1962, Varley and Moroz (14) conducted studies of yeasts isolated from various refinery materials. These investigators did not report isolating Hansenula species. Tilbury (13) in a study of 96 cultures of yeast isolated from raw sugars classified six of these isolates in the Hansenula genus. Sherwood and Hines (10) reported isolating Hansenula anomala in several lots of deteriorating raw sugars. Recently, Scarr (9) reported an increase in numbers of Hansenula anomala and Torulopsis species in various samples of raw sugars examined at the Tate and Lyle Ravensbourne Research Center.

An increase in numbers of Hansenula yeasts received in shipments of raw sugars could possibly have deleterious effects in sugar refining. These yeasts possess extremely active enzyme systems and, under certain conditions, can produce appreciable amounts of extra-cellular invertase. Also, acetic acid is produced in large quantities by certain species of Hansenula, such as Hansenula anomala, but the acetic acid is enzymatically bound to ethanol to produce ethyl acetate. Several species in the genus Hansenula can form highly viscous solutions of phosphomannans. This extracellular capsular polysaccharide contains only mannose in addition to phosphate.

Therefore, because of the highly developed enzyme systems of these yeasts and their possible implications in the overall biological sucrose loss program, a study was undertaken to determine the occurrence of Hansenula yeasts in raw sugars.

Our preliminary survey showed that, of a total of 265 samples of raw sugars examined for the presence of Hansenula yeasts, 145 samples gave a positive determination for Hansenula yeasts (table 10). In this preliminary survey a positive determination indicated that at least one Hansenula yeast was present in the 30 gram sample of raw sugar examined. No attempt was made in this survey to determine an actual count of Hansenula yeast present in each sample of raw sugar examined. A subsequent detailed examination of four lots of raw sugars showed that 12% of the yeast enumerated from positive raw sugar samples was tentatively identified as Hansenula yeast (table 11). Since the criterion for identification was the single physiological characteristic of nitrate assimilation, the relative proportion of Hansenula yeasts in the four lots of raw sugar were only approximately related to the number of yeasts enumerated in each lot of raw sugar.

Six cultures of yeast, isolated from a Mexican raw sugar, were positively identified as Hansenula species.

TABLE 10--Hansenula/nitrate reducing yeasts survey of raw sugars

Country of origin	Number of lots examined	Number of samples examined	Positive determination Hansenula/nitrate reducing yeast
Peru	3	83	43
Philippine Islands	5	89	72
Mexico	2	28	14
Brazil	3	9	3
Australia	1	30	6
Puerto Rico	2	9	3
Dom. Republic	2	17	4
	18	265	145

TABLE 11--Determining number of Hansenula yeast in four lots of raw sugar

	Description of samples	Preliminary determination results recorded per 30 grams of raw sugar	Total yeast count in positive samples recorded per 10 grams	Tentatively ident. as Hansenula
1.	Australia - Lot #908 - Philippines	+	50	13
2.	" " "	-	-	-
3.	" " "	-	-	-
4.	" " "	-	-	-
5.	" " "	-	-	-
6.	" " "	+	37	4
7.	" " "	-	-	-
8.	" " "	-	-	-
1.	Philippine Island-Lot #907-Brooklyn	-	-	-
2.	" " "	-	-	-
3.	" " "	+	80	17
4.	" " "	+	20	2
5.	" " "	-	-	-
6.	" " "	-	-	-
7.	" " "	-	-	-
8.	" " "	+	60	11
1.	Mexican - Lot #953 - Boston	-	-	-
2.	" " "	+	50	4
3.	" " "	-	-	-
4.	" " "	+	40	2
5.	" " "	+	28	3
6.	" " "	-	-	-
7.	" " "	-	-	-
8.	" " "	-	-	-
1.	Dominican Republic - Lot #939 - Boston	-	-	-
2.	" " " "	-	-	-
3.	" " " "	+	120	8
4.	" " " "	-	-	-
5.	" " " "	-	-	-
6.	" " " "	+	35	10
7.	" " " "	+	5	1
8.	" " " "	+	10	2

TABLE 12--Invertase activity of Hansenula yeast species compared to Saccharomyces yeast species.

	Invertase activity, units per ml of culture broth
Hansenula ASC 2-A	2,960
Hansenula ASC 2-B	2,000
Hansenula ASC 4-A	3,190
Hansenula ASC 4-B	2,280
Hansenula ASC 5-A	2,090
Hansenula ASC 5-B	1,950
Saccharomyces Species*	0
Saccharomyces Species*	1,130

*Yeast recorded in tables 1 to 3

Several years ago, Dworschack and Wickerham (4) conducted a survey to determine the production of extracellular invertase of 68 yeast species involving 17 different genera. These investigators reported the highest yield of extracellular invertase was produced by Hansenula jadinii. Employing the Dworschack and Wickerham technique for invertase activity, a study was conducted comparing the six Hansenula isolates to various Saccharomyces species involved in previous invertase activity studies. The data recorded in table 12 showed the extremely high levels of invertase production of the Hansenula cultures, ranging from 1950 units to 3190 units. This high range of invertase activity compares to the Saccharomyces species range of 0 units to 1130 units.

MOLDS

Molds are minute saprophytic or parasitic filamentous fungi which reproduce by means of asexual or sexual spores. Molds possess a thallus which consists of branching, threadlike growth called hyphae. A mass of threads taken collectively is spoken of as a mycelium.

Molds are especially characterized by the ability to elaborate a great variety of enzymes and this physiological qualification undoubtedly accounts for their ability to thrive on so many materials and in the presence of very small amounts of organic matter.

In contrast to yeasts, it was possible to estimate fairly accurately the relative frequency of occurrence of the various mold species. Differences in the colonial morphology between species were more apparent and identification was more rapid.

In a taxonomic study involving 467 samples of offshore and Louisiana raw sugars we found the most common genus to be Aspergillus, with smaller numbers of

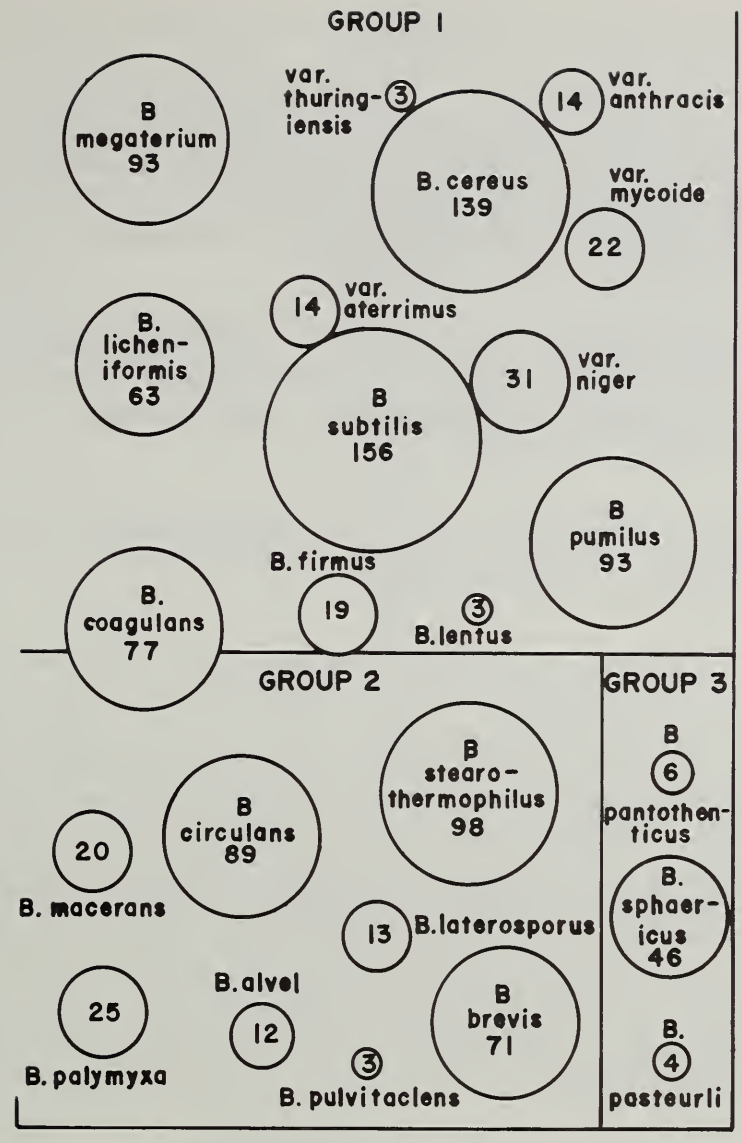

FIGURE 1--The classification and possible natural relationship of Bacillus species.

Group 1 - Sporangia not definitely swollen
Group 2 - Sporangia definitely swollen by oval spores
Group 3 - Sporangia swollen by round spores

From: Smith, N. R., Gordon, R. E., and Clark, F. E. (1952) Aerobic Sporeforming Bacteria Agric. Monogr., U.S. Dept. Agric. No. 16

Penicillium and other genera. These mold species were identified to the genus level by reference to Thom and Raper (12), Raper and Thom (7), and Barnett (1).

BACTERIA

Bacteria are defined as microscopic single-celled plants that usually reproduce by binary fission, the simplest form of reproduction known. Over 1600 species of bacteria have been studied and classified. These represent probably only a small percentage of the total number of species in existence. Since bacteria are so small and undifferentiated, it is an exceedingly difficult matter to identify and classify them.

The majority of the bacteria surviving the refining process or found in raw sugar appear to be spore-forming bacteria. On occasion, non-spore forming bacteria such as the Leuconostocs, Flavobacterium, Aerobacters, and Lactobacilli have been isolated from sugar products.

There are two genera comprising the family Bacillaceae (spore-forming rods). Bacillus species are aerobic or facultative, Clostridium species are anaerobic. Most species are saprophytic soil organisms.

Smith, Gordon, and Clark (11) studied extensively 1134 strains of aerobic spore-formers and clarified classification considerably. They used both morphological criteria (shape and location of the spore in the cell, shape of the sporangia, diameter of the cell, etc.) and physiological criteria (acid production from various carbohydrates, hydrolysis of gelatin, starch, etc.). Of 175 species names previously proposed, the authors retained 22. Their classification was also a first attempt to introduce natural relationships amongst the species. The main emphasis was on the morphology of the organisms. Within these arbitrary limits, species differentiation was based on the greatest number of commonly occurring properties. The classification is shown in figure 1. The present studies of aerobic spore-formers followed closely the materials and methods as outlined by Smith, Gordon, and Clark (11) with final identification made by reference to Bergey (2).

Aerobic Thermophilic Bacteria

As a part of a taxonomy program on aerobic thermophilic bacteria, 641 representative colonies were isolated from 143 samples of white granulated sugars. In developing a taxonomic profile on these cultures the following results were obtained:

Since this particular study dealt with "flat-sour" organisms, all 641 cultures were spore-forming, and were grown at 55°C incubation. The first study involved an attempt to group these 641 cultures according to temperature for growth. The results of this study indicated two major groups. Group No. 1 was composed of 196 cultures which grew only at 55°C and not at 28°C or 37°C. Group No. 2 was composed of 445 cultures which grew at 28°C, 37°C, and 55°C.

Representative cultures obtained from each group were identified according to Bergey (2). Group No. 1 was a pure culture of Bacillus stearothermophilus while group No. 2 was heterogeneous group composed of four members of the genus Bacillus: Bacillus brevis, Bacillus coagulans, Bacillus circulans and Bacillus subtilis.

TABLE 13--Carbohydrate fermentation patterns of 38 bacteria isolated from raw sugars and finished products, employing fourteen carbohydrate sources incorporated in two base nitrogen media

Carbohydrate Tested	Percentage of Organisms showing positive fermentation in base nitrogen media	
	Organic	Inorganic
Sucrose	100	100
Dextrose	100	100
Fructose	100	100
Salicin	89	66
Maltose	87	87
Galactose	79	71
Mannose	55	47
Glycerol	42	66
Arabinose	13	3
Lactose	13	0
Mannitol	13	18
Sorbitol	11	8
Xylose	8	3
Rhamnose	3	0

Aerobic Mesophilic Bacteria

Considerable data had been accumulated on numerous morphological and physiological tests performed on approximately 250 cultures of aerobic mesophilic spore-forming bacteria.

All 250 cultures grew well between 25°C and 40°C with optimal growth between 28°C to 32°C. Approximately 75% of the cultures were represented by Bacillus megaterium, Bacillus subtilis, Bacillus cereus, and Bacillus brevis. The remaining 25% were represented by Bacillus circulans, Bacillus sphaericus, Bacillus coagulans, and Bacillus polymyxa.

Many different types of carbohydrates, ranging from the simple sugars to more complex carbohydrates, may be fermented by bacteria. In a carbohydrate profile, this laboratory uses 14 different carbohydrate sources, recording both gas and acid production. The 14 carbohydrate sources are incorporated into 2 nitrogen base media. The first base medium, Cystine Trypticase Agar, makes use of the amino acid cystine, and peptone trypticase, derived from casein by pancreatic digestion, as the principal sources of nitrogen. In the second base medium, Ayers, Rupp, and Johnson Medium, the inorganic compound dibasic ammonium phosphate is used as the principal source of nitrogen.

Table 13 shows the number of positive acid production reactions obtained by testing 38 representative cultures of aerobic mesophilic spore formers. In this study, all the 38 cultures showed negative gas production. These 38 cultures, representing isolates from various raw sugars and finished products, both liquid

and granulated, followed closely the patterns established in previous carbohydrate profile studies of the spore-forming organisms. All cultures fermented glucose, fructose, and sucrose, with varying positive reactions with the other carbohydrate sources. There was very little difference in the number of positive responses utilizing the two nitrogen bases. In the case of the carbohydrates glucose, fructose, and sucrose, the organisms were able to ferment these carbohydrates regardless of the nitrogen sources.

Bacteria vary widely in their ability to utilize the various sources of nitrogen for the synthesis of protein. However, many species of bacteria may utilize nitrate as the sole source of nitrogen, probably first reducing the nitrate to nitrite, then to hydroxylamine, and finally to ammonia in the process of assimilation. The ability of bacteria to reduce nitrate ion to nitrite or to ammonia is measured as one of the physiological characteristics for the differentiating of bacteria.

The 38 organisms in this study all gave a positive test for nitrate reduction.

Anaerobic Thermophilic Bacteria

Identification of this class of microorganisms were restricted to the "sulfur-stinkers" (Clostridium nigrificans) and anaerobic gas formers (Clostridium thermosaccharolyticum).

Anaerobic Mesophilic Bacteria

Through the years, organoleptic controls on the odor quality of certain sugars indicate the occasional presence of an objectional odor attributable to trace quantities of butyric acids. This acid produces a characteristic odor detectable by the human olfactory system in quantities as small as one part per million.

Preliminary investigation of sugars evincing butyric acid odors resulted in the isolation of a spore-forming anaerobic bacteria identified as Clostridium butyricum. This investigation also showed that this organism was capable of fermenting lactate to butyric acid.

Other Bacteria of Special Interest

The presence of polysaccharides was first recognized in the sugar industry many years ago. During the latter part of the 19th century, dextran was observed in sugar refineries as masses of slime that attracted attention because they impeded purification of sugar.

A study was conducted to enumerate the aerobic spore-formers found in offshore and Louisiana raw sugars that possess the ability to synthesize polysaccharides from a sucrose source. From 467 test samples, 57 representative cultures were isolated in pure culture and identified according to Bergey (2). An evaluation of results recorded from over 70 morphological and physiological tests showed that 40 cultures were identified as Bacillus subtilis, and 17 cultures identified as Bacillus cereus.

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E. Boloker (Amstar): What primary isolation medium did you use to isolate yeast cultures in your study?

R. D. Skole: As yeasts are capable of development over a fairly wide Brix range, their isolation normally presents few if any difficulties. However, we were primarily interested in isolating the yeast species classified as osmophilic or osmotolerant. Therefore, we utilized the yeast-malt extract agar with 30% added sucrose that was devised by Dr. Wickerham[1] at NRRL[2] for isolating yeasts from substrates of high osmotic pressure.

R. S. Patterson (C&H): What temperature is needed to destroy the spores?

R. D. Skole: The heat resistance of microorganisms usually is expressed in terms of their thermal death time, which is defined as the time it takes at a certain temperature to kill a stated number of spores or organisms under specified conditions. The heat resistance of bacterial spores varies greatly with the species and conditions during sporulation. In the microbiological laboratory, the usual sterilizing agent is steam at a temperature of 121°C. To secure this temperature, the steam must be under pressure, approximately 15 lb per sq in. Exposure for 15 min is sufficient to kill all bacterial spores.

J. C. P. Chen (Southdown): We have been talking about the impurities in raw sugar produced from cane. Do we know anything about how much of this is caused by microorganisms? We have heard that the cane produces a polysaccharide and a protein that are implicated in floc formation. We think that a lot of the dextran comes from Leuconostoc, but we have heard that dextran does not cause floc formation. How much of these floc formers come from microorganisms?

R. D. Skole: Several years ago we conducted a survey using a direct microscopic stain devised by Coca-Cola. This differentiated between viable and nonviable yeast cells. It gave us a number count of yeast cells. In certain raw sugars the viable count was about two hundred, but the number of dead yeast cells was millions, even in the billions, per gram. These yeast cells contain a certain amount of protein, which would show up as protein in the raw sugar.

R. Moroz (SuCrest): Have you ever found any Salmonella or Staphylococcus

R. D. Skole: No. We have had an ongoing microbiological survey program at our Research and Development Center for at least 10 years; it's a very comprehensive program. To date, we have not enumerated any Salmonella.

R. Moroz: Does Salmonella grow in sugar solutions?

R. D. Skole: As a rule, relatively low concentrations of sucrose (from 1 to 20 Brix) will stimulate microbial growth, while higher concentrations of sucrose inhibit them. The rate of growth of Salmonella or any other species of bacteria in substrates containing sucrose or other solutes is a function of the

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[1] See Reference No. 16.
[2] Northern Regional Research Center, ARS-USDA, Peoria, Illinois.

water activity (a_w) and generally not of the concentration of a particular solute or solutes. As the solute concentration is increased, the water activity is decreased. With a decreased a_w, the lag phase of the organism increases, and the rate of growth decreases. From a practical point of view, application of water activity principles means that the various grades of liquid sugars will not support the growth of Salmonella or other pathogens.

R. Moroz: Do you need some protein to develop Salmonella?

R. D. Skole: Yes.

N. H. Smith (C&H): Does table 13 show products of bacterial action?

R. D. Skole: These are products of the polysaccharide formed by bacterial action.

N. H. Smith: And these include the same kind of things - uronic acids and various sugars - that ISP has. Since bacterial action predominates more at low Brix and ISP is found in cane juice sometimes and not other times, what is the possibility that ISP is formed by bacterial action? Do these bacteria grow fast enough that they could produce ISP?

R. D. Skole: The population of a normal bacterial culture doubles in each consecutive time interval, so that the number of bacteria increases in an exponential or logarithmic progression. The rate of cell division can be expressed in various ways. It is most commonly stated as the generation time, the interval required for one generation. The multiplication rate of bacteria varies from one species to another.

Rapidly dividing bacteria like E. coli have a minimum generation time of 13 to 17 min. Therefore, under certain growth conditions, bacterial growth rate can be quite rapid.

CODEX STANDARDS FOR SUGARS

By John A. Hupfer[1]

ABSTRACT

The work of the Codex Alimentarious Commission's Committee on Sugars in developing international (Codex) standards for sugars is discussed in the light of the United States participation in this activity. Standards prepared for 10 carbohydrate sweeteners are reviewed, including White Sugar, Powdered Sugar and Soft Sugars. The current status of each standard and the progress made toward accepting Codex standards in this country are noted.

INTRODUCTION

The first step toward developing international standards for foods and food products was taken in 1962 at a joint conference on Food Standards held by the Food and Agriculture Organization (FAO) of the United Nations and the World Health Organization (WHO). These organizations decided to establish the Codex Alimentarious Commission, the agency responsible for the work of developing international food standards. The Commission held its first session in 1963. Eleven sessions of the Commission have been held to date. Membership in the Commission has grown stadily from 50 countries to a current membership of 114 countries.

The basic objectives of the Commission are to protect the interests of consumers and to facilitate world trade. International (Codex) standards for foods contain requirements aimed at ensuring for the consumer a sound, wholesome food product free from adulteration, correctly labelled and presented.

Most of the work of the Commission is carried on through committees. Commodity committees were set up to draft standards for various foods. General subject committees were organized to prepare requirements for general application to food standards as for example, in food hygiene, food labelling, methods of analysis and sampling, etc.

The Codex Committee on Sugars was formed in 1964 and given responsibility for drafting standards for carbohydrate sweeteners with the exception of honey. All six sessions of the Committee have been hosted by the United Kingdom in London. Five of the sessions were held in the years 1964 through 1968. The sixth and latest session took place in 1974.

The Committee drafted standards for ten carbohydrate sweetener products. These include White Sugar, Powdered Sugar, Soft Sugars, Glucose Sirup, Dried Glucose Sirup, Dextrose Monohydrate, Dextrose Anhydrous, Powdered Dextrose, Lactose and Fructose. Those sections of the standards which relate to the scope of the standard (if included), product description, essential composition and

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[1]U.S. Representative to the Codex Committee on Sugars, U.S. Department of Agriculture, ASCS-Commodity Operations Division, Washington, D.C. 20250.

quality factors, food additives, and contaminants are set forth for each of these products in appendices 1 through 10, respectively.

Other requirements in the standards set forth in sections related to hygiene, labelling, and methods of analysis and sampling are, for the most part, not discussed in this paper and are not included in the appendices. A discussion of each of these sugars and the standards which have been prepared for them follows:

WHITE SUGAR

There are three principal types of white sugars: a highly purified white granulated sugar, plantation white granulated sugar and turbinado sugar. The pol averages about 99.9°S for highly purified white sugar, 99.7°S for plantation white sugar and 99.5°S for turbinado sugar. Poorer keeping quality, color, and taste characteristics of plantation white and turbinado sugars severely limit their use in industry and their acceptability on the part of consumers, particularly in developed countries. The great bulk of the sugar production in this country and other developed, technologically advanced countries consists of highly purified white sugar, whereas developing countries manufacture significant quantities of plantation white granulated, turbinado, and other types of direct consumption sugars.

There was considerable discussion in Committee during its first four sessions on the issue of whether to include all types of white sugars under a single standard or to prepare separate standards for white sugars. A majority of the delegates were in agreement that a single standard would be adequate for consumer protection and best meet the objectives of the Codex. However, a substantial minority took the view that the number of standards should be based on international trade practices. With all three types of white sugars moving in international commerce, the minority contended that a three standard system should be developed for the sake of consumer protection.

The Committee decided to prepare a single white sugar standard with two separate specifications. Requirements in Specification A are applicable to highly purified white sugar whereas those in Specification B apply to lower purity plantation white and turbinado sugars. The standard appears in Appendix 1.

Labelling requirements in the standard require that all products designated as white sugar without qualification must conform to Specification A. Products conforming to Specification B but not conforming to Specification A may be designated plantation white sugar, mill white sugar or any equivalent name accepted for such sugar in the country in which it is sold, or any designation which is not misleading and which does not include the word white.

This standard was advanced step by step through the Codex procedure for the elaboration of world-wide standards. It was accepted by the Commission as recommended standard in 1969 and formally presented to our government for acceptance in January 1971. It is still under consideration.

POWDERED SUGAR

Powdered sugar (icing sugar) is finely pulverized white sugar with or without the addition of an anti-caking agent. The standard for this product is set forth in Appendix 2.

Essential composition and quality factors in the standard relate to that part of the powdered sugar other than the anti-caking agent or agents. The minimum requirements and maximum limits placed on the compositional and quality factors are the same as those for white sugar that conforms to Specification A of the White Sugar Standard.

The standards for white sugar and powdered sugar were advanced together through the Codex procedure for elaborating worldwide standards. The recommended standard for powdered sugar has been subject to consideration by the U.S. government since 1971.

In 1974, the Committee was informed that methods of analysis could not be provided for the enforcement of the compositional and quality criteria in the standard because of interference caused by the presence of anti-caking agents. The Committee agreed to reconsider the standard for the purpose of setting new quality requirements based entirely on the final product sold rather than upon the white sugar used to make the product.

A considerable amount of work will have to be accomplished before the Committee can work out the details for the prospective change. Both the Committee and the Commission have agreed that the changes contemplated in the standard would be accomplished by amendment.

SOFT SUGARS

At its second sesion in March 1965, the Committee decided to propose a single standard for soft sugars to the Commission for adoption as a draft provisional standard. The Commission accepted the proposal and submitted the draft provisional standard to governments for comment.

The United States recognized that a wide variety of sugars are manufactured which may be properly classified as soft sugars. These sugars range from soft white sugar to dark brown sugar and differ considerably in color, flavor, composition, and uses. In view of this, the United States took the position that separate standards should be prepared for the two major categories of soft sugars i.e., soft white sugar and soft brown sugars. Further, the variations existing among soft brown sugars were considered sufficient to warrant separate specifications designating some of the principal types of brown sugars. Accordingly, the United States proposed definitions and specifications for a standard for soft white sugar and a standard for soft brown sugars, containing specifications for light brown sugar, medium brown sugar and dark brown sugar.

The Committee considered the comments made by governments on the draft provisional standard at the fourth session, in 1967. The Committee did not accept the U.S. representatives proposal for establishing separate standards for these products. However, the Committee did decide to draw distinction between soft

white sugar and soft brown sugars by adding a special specification within the standard for soft sugars to which products labelled soft white sugar must comply.

In view of extensive changes made to the draft provisional standard at the fourth session, the Committee decided to circulate the standard in its amended form to governments for comment rather than propose the standard to the Commission for adoption as a recommended standard. The Committee considered the comments received from governments at its fifth session, in 1968, and proposed the standard for adoption as a recommended Codex standard. The Commission adopted the standard in 1969 and presented the recommended Codex standard for soft sugars to the U.S. government for acceptance in 1971. To date no decision has been made by the U.S. government with regard to accepting this standard. The standard for soft sugars is in Appendix 3.

GLUCOSE SIRUPS AND DEXTROSE

At the first session of the Committee, in March 1964, standards were drafted for four sugars which are derived from starch, namely: glucose sirup, dried glucose sirup, dextrose monohydrate, and dextrose anhydrous. All of these products are made from corn starch in the United States. Glucose sirup is commonly referred to in the United States as corn sirup.

The Committee modified the standards at its Second Session, in 1965, and proposed them to the Commission as draft provisional standards. The Commission accepted the Committee's proposals as draft provisional standards and sent them to governments for comment.

The Committee considered the comments received from governments at its Fourth Session, in May 1967. The standards were amended and advanced to the Commission for adoption as recommended standards. The Commission adopted the standards for all four sugars, as recommended standards in 1968.

Recommended standards for glucose sirup, dried glucose sirup, dextrose monohydrate, and anhydrous dextrose are in Appendices 4, 5, 6, and 7, respectively. These standards were formally presented to the U.S. government for acceptance in January 1971. These standards were accepted by the U.S. government in 1974 and promulgated as standards of identity under the authority of the Federal Food, Drug and Cosmetic Act. The U.S. standards of identity for these sugars are identical to the Codex standards in most respects.

POWDERED DEXTROSE

A standard for powdered dextrose was drafted on the lines of the standard already prepared for Powdered Sugar. The draft standard for powdered dextrose was adopted as a recommended standard at the Eighth Session of the Codex Commission, in July 1971. The standard is in Appendix 8. It is still under consideration for acceptance in the United States.

Provision is made in the standard for the use of the same anti-caking agents, in like amounts, as specified in the recommended standard for powdered

sugar. In addition, the quality criteria in the recommended standard for powdered dextrose are also based on the dextrose used to make the product rather than on the product as sold. Consequently, the Committee is confronted with the same problems arising from the interference caused by the presence of anti-caking agents on analytical test results as were noted earlier for the Powdered Sugar Standard. In this case, the Committee also intends to place the quality requirements on the basis of the product as sold. Necessary changes in the standard will be accomplished by amendment.

LACTOSE

A standard for lactose was drafted at the Second Committee Meeting, in 1965, and sent on to the Commission for adoption as a draft standard. The standard was accepted by the Commission and circulated to governments for comment.

The United States considered that some of the analytical criteria in the standard were too restrictive. The criteria were considered better suited to a pharmaceutical grade lactose than to an edible grade of lactose for food use.

The United States had strong feelings about the matter and presented a paper setting forth its views and recommendations for consideration by delegates to the Third Session of the Committee, in March 1966. The Committee revised the standard to conform with most of the U.S. recommendations before submitting it to the Commission. The Commission accepted the standard as a draft provisional standard in November 1966.

The standard was revised further to our satisfaction at the Fourth Session of the Committee, in May 1967. The standard was proposed to and accepted by the Commission as a recommended standard in 1968.

The Recommended Standard for Lactose, which appears in Appendix 9, was presented to the U.S. government for acceptance in January 1971. The standard was accepted by the United States in 1975 and promulgated as a standard of identity under the Food, Drug and Cosmetic Act.

FRUCTOSE

The Committee decided in 1965 to prepare a standard for fructose, largely on the strength of a recommendation made by Germany, a major producer of the product. In 1966, Germany withdrew its recommendation. This action, together with the small quantities of fructose that were being traded internationally, led to a decision on the part of the Committee to discontinue the work of developing the standard.

Prior to the Sixth Session of the Committee in March 1974, another Codex group working on the standardization of fruit juices requested the Sugars Committee to consider preparing a standard for fructose. They noted that fructose had been added to the list of sugars in some of their standards.

In order to provide a basis for deciding this issue, a survey was made in advance of the meeting to obtain production, consumption, import, and export statistics for fructose. At that time, the world production was estimated between 30 and 40 thousand tons annually. Quantities of fructose moving in international trade were still small. The Committee noted, however, that in recent years fructose production has been rising due to its increasing use as a food ingredient. The product has been gaining increasing recognition as a sweetener with a wide range of potential applications when used alone or in combination with other sweeteners.

The Committee recognized the growing importance of the product and decided to develop a standard for fructose which would be suitable for application to foods as distinct from pharmaceuticals. A proposed draft standard was sent out to governments for comment during the second half of 1974.

Rather than call a special session of the Committee to consider the government comments, the Committee Secretariat considered the comments and prepared a revision of the proposed draft standard for consideration by the Codex Commission at its Eleventh Session, held in April 1976. The Commission accepted the standard which the Secretariat had revised in line with our recommendations. The draft standard for fructose is set out in Appendix 10.

At this point, it appears that this standard will be advanced steadily and presented to the U.S. government as a recommended standard in due course.

Appendix 1.--International Standard for White Sugar

1. Scope

This standard applies to white sugar except that paragraph 3.1.4 (Loss on drying) does not apply to white sugar in lump or cube form or to crystal candy sugar (crystal korizato) or to rock sugar (korizato).

2. Description

White sugar is purified and crystallized sucrose (saccharose).

3. Essential Composition and Quality Factors

3.1 Specification A

3.1.1 Polarization	not less than 99.7°S
3.1.2 Invert sugar content	not more than 0.04% m/m (mass/mass)
3.1.3 Conductivity ash	not more than 0.04% m/m
3.1.4 Loss on drying (3 hours at 105°C)	not more than 0.1% m/m

3.1.5 Color	not more than 60 ICUMSA units
3.2 Specification B	
3.2.1 Polarization	not less than 99.5°S
3.2.2 Invert sugar content	not more than 0.1% m/m
3.2.3 Conductivity ash	not more than 0.1% m/m
3.2.4 Loss on drying 3 hours at 105°C	not more than 0.1% m/m
3.2.5 Color	not more than 150 ICUMSA units

4. Food Additives

4.1 Sulfur dioxide	
Specification A	not more than 20 mg/kg
Specification B	not more than 70 mg/kg

5. Contaminants

Specifications A and B	
5.1 Arsenic (As)	not more than 1 mg/kg
5.2 Copper (Cu)	not more than 2 mg.kg
5.3 Lead (Pb)	not more than 2 mg/kg (temporarily endorsed)

NOTE: Sections 6, 7, and 8 are omitted from this Appendix. These sections are: Hygiene, Labelling, and Methods of Analysis and Sampling.

Appendix 2.--International Standard for Powdered Sugar (Icing Sugar)

1. Description

Powdered sugar (icing sugar) is finely pulverized white sugar with or without the addition of an anti-caking agent.

2. Essential Composition and Quality Factors

Optional ingredients

Starch	not more than 5% m/m of the product provided that no other anti-caking agent is used.

Quality criteria

That part of powdered sugar, other than the anti-caking agent or agents, shall conform to the following specifications:

Polarization	not less than 99.7°S
Invert sugar content	not more than 0.04% m/m
Conductivity ash	not more than 0.04% m/m
Loss on drying (3 hours at 105°C)	not more than 0.1% m/m
Color	not more than 60 ICUMSA units

3. Food Additives

Sulfur dioxide (residue resulting from the white sugar used)	not more than 20 mg/kg

Anti-caking agents

The following may be used, singly or in combination, provided that starch is not present:

Calcium phosphate, tribasic Magnesium carbonate Magnesium stearate Silicon dioxide, amorphous (dehydrated silica gel)	Not more than 1.5% m/m

Silicates:	)
Calcium silicate	)
Magnesium trisilicate	)
Sodium calcium alumino-silicate	)

4. Contaminants

Arsenic (As)	not more than 1 mg/kg
Copper (Cu)	not more than 2 mg/kg
Lead (Pb)	not more than 2 mg/kg (temporarily endorsed)

NOTE: Sections 5, 6, and 7 are omitted from this Appendix.

Appendix 3.--International Standard for Soft Sugars

1. Description

The term soft sugar means fine-grain purified moist sugar and does not include large-grain brown or yellow sugars such as Demerara sugar.

2. Essential Composition and Quality Factors

Specification A: Soft Sugar (other than White Soft Sugar)

Sucrose (saccharose) plus invert sugar content	not less than 88.0% m/m expressed as sucrose
Invert sugar content	not less than 0.3% m/m, not more than 12.0% m/m
Sulfated ash	not more than 3.5% m/m
Loss on drying (3 hours at 105°C)	not more than 4.5% m/m
Color	white to dark brown

Specification B: White Soft Sugar

Sucrose (saccharose) plus invert sugar content	not less than 97.0% m/m expressed as sucrose
Invert sugar content	not less than 0.3% m/m not more than 12.0% m/m
Conductivity ash	not more than 0.2% m/m
Loss on drying (3 hours at 105°C)	not more than 3.0% m/m
Color	not more than 60 ICUMSA units

3. Food Additives

Sulfur dioxide	not more than 40 mg/kg

4. Contaminants

Arsenic (As)	not more than 1 mg/kg
Copper (Cu)	not more than 10 mg/kg
Lead (Pb)	not more than 2 mg/kg (temporarily endorsed)

NOTE: Sections 5, 6, and 7 are omitted from this Appendix.

Appendix 4.--International Standard for Glucose Sirup

1. Description

Glucose sirup is a purified concentrated aqueous solution of nutritive saccharides obtained from starch.

2. Essential Composition and Quality Factors

Total solids content	not less than 70.0% m/m
Reducing sugar content (Dextrose equivalent)	not less than 20.0% m/m, expressed as D-glucose, on a dry basis
Sulfated ash	not more than 1.0% m/m, on a dry basis

3. Food Additives

Sulfur dioxide	not more than 40 mg/kg
Sulfur dioxide in glucose sirup for the manufacture of sugar confectionery only	not more than 400 mg/kg

4. Contaminants

Arsenic (As)	not more than 1 mg/kg
Copper (Cu)	not more than 5 mg/kg
Lead (Pb)	not more than 2 mg/kg (temporarily endorsed)

NOTE: Sections 5, 6, and 7 are omitted from this Appendix.

Appendix 5.--International Standard for Dried Glucose Sirup

1. Description

Dried glucose sirup is glucose sirup from which the water has been partially removed.

2. Essential Composition and Quality Factors

Total solids content	not less than 93.0% m/m

Reducing sugar content (Dextrose equivalent)	not less than 20.0% m/m expressed as D-glucose, on a dry basis
Sulfated ash	not more than 1.0% m/m on a dry basis

3. Food Additives

Sulfur dioxide	not more than 40 mg/kg
Sulfur dioxide in dried glucose sirup for the manufacture of sugar confectionery only	not more than 150 mg/kg

4. Contaminants

Arsenic (As)	not more than 1 mg/kg
Copper (Cu)	not more than 5 mg/kg
Lead (Pb)	not more than 2 mg/kg (temporarily endorsed)

NOTE: Sections 5, 6, and 7 are omitted from this Appendix.

Appendix 6.--International Standard for Dextrose Monohydrate

1. Description

Dextrose monohydrate is purified and crystallized D-glucose containing one molecule of water of crystallization per glucose molecule.

2. Essential Composition and Quality Factors

D-glucose content	not less than 99.5% m/m, on a dry basis
Total solids content	not less than 90.0% m/m

Sulfated ash	not more than 0.25% m/m, on a dry basis

3. Food Additives

Sulfur dioxide	not more than 20 mg/kg

4. Contaminants

Arsenic (As)	not more than 1 mg/kg
Copper (Cu)	not more than 2 mg/kg
Lead (Pb)	not more than 2 mg/kg (temporarily endorsed)

NOTE: Sections 5, 6, and 7 are omitted from this Appendix.

Appendix 8.--Powdered Dextrose (Icing Dextrose)

1. Description

Powdered dextrose (icing dextrose) is finely pulverized dextrose anhydrous or dextrose monohydrate or mixtures thereof, with or without the addition of an anti-caking agent.

2. Essential Composition and Quality Factors

Optional Ingredients

Starch	not more than 5% m/m of the product, provided that no other anti-caking agent is used.

Quality Criteria

That part of powdered dextrose, other than the anti-caking agent or agents, shall conform to the following specifications:

Reducing sugar (dextrose equivalent)	not less than 99.5% m/m on a dry basis

Total Solids Content

Powdered dextrose made from dextrose anhydrous: not less than 98.0% m/m

Powdered dextrose made from dextrose monohydrate: not less than 90.0% m/m

Powdered dextrose made from dextrose anhydrous or dextrose monohydrate or mixtures thereof: the total solids content shall be proportional to the characteristics of the mixture.

Sulfated ash	not more than 0.25% m/m on a dry basis

3. Food Additives

Sulfur dioxide (residue resulting from the dextrose used)	not more than 20 mg/kg

Anti-caking agents

The following may be used, singly or in combination, provided that starch is not present:

Calcium silicate Calcium phosphate, tribasic Magnesium carbonate Magnesium stearate Silicon dioxide, amorphous (dehydrated silica gel) Silicates: Magnesium trisilicate Sodium calcium aluminosilicate	not more than 1.5% m/m

4. Contaminants

Arsenic (As)	not more than 1 mg/kg
Copper (Cu)	not more than 2 mg/kg
Lead (Pb)	not more than 2 mg/kg

NOTE: Sections 5, 6, and 7 are omitted from this Appendix.

Appendix 9.--International Standard for Lactose

1. Description

Lactose is the carbohydrate normally obtained from whey. It may be anhydrous or contain one molecule of water of crystallization per molecule of lactose, or be a mixture of both forms.

2. Essential Composition and Quality Factors

Anhydrous lactose content	not less than 99.0% m/m, on a dry basis
Sulfated ash	not more than 0.3% m/m, on a dry basis
Loss on drying (16 hours at 120°C)	not more than 6.0% m/m
pH (Solution 10% m/m)	4.5 - 7.0

3. Food Additives

None permitted.

4. Contaminants

Arsenic (As)	not more than 1 mg/kg
Copper (Cu)	not more than 2 mg/kg
Lead (Pb)	not more than 2 mg/kg (temporarily endorsed)

NOTE: Sections 5, 6, and 7 are omitted from this Appendix.

Appendix 10.--Draft Standard for Fructose

1. Description

Fructose is purified and crystallized D-fructose.

2. Essential Composition and Quality Factors

Specific Rotation $[\ ]_D^{20}$ (10 g/100 ml)	-89° to -93.5°
Loss on drying (5 hours at 70°C)	not more than 0.5% m/m
Sulfated Ash	not more than 0.1% m/m

Color	not more than [60] ICUMSA units
pH (solution 10% m/m)	4.5 - 7.0

3. Food Additives

Sulfur dioxide (residue from raw material)	not more than 20 mg [kg]

4. Contaminants

Arsenic (As)	not more than 1 mg/kg
Copper (Cu)	not more than 2 mg/kg
Lead (Pb)	not more than 2 mg/kg

NOTE: Sections 5, 6, and 7 are omitted from Appendix.

DISCUSSION

K. R. Hanson (Amstar): If we were to read in tomorrow morning's newspaper that the Food and Drug Administration had accepted the Codex standard for white sugar, what would that mean to us as refiners?

J. A. Hupfer: It would mean that if you sold sugar labeled as white sugar, it would have to meet the requirements of the standard.

K. R. Hanson: Does this include all sugar that is sold internally in the United States?

J. A. Hupfer: Yes, the Codex standard would be applicable to all white sugar marketed in the United States, regardless of whether it was internally produced or imported.

K. R. Hanson: Would this then become a Standard of Identity?

J. A. Hupfer: If the recommended standard for white sugar is accepted by the U.S. government, it would be promulgated as a standard of identity by the Food and Drug Administration (FDA). The same procedure would be followed in promulgating this international (Codex) standard as would be followed in the case of a domestic food standard. The general principles of the Codex Alimentarius provide that countries may accept standards in accordance with their own existing laws and regulations for establishing food standards.

As to whether countries incur any obligation to accept Codex standards through participation in the work of the Codex Alimentarious, the Codex

Alimentarius Commission has no authority to force acceptance of its standards on the part of any government, including those that have taken part in developing the standards. Countries are free to either accept or reject Codex standards.

J. C. P. Chen (Southdown): So far we have been using Bottlers' Standards for refined sugar and liquid sugar. I know we don't have a national standard for the U.S. We don't have a national standard for arsenic, copper, and lead. If the United States accepted the International Codex standard, will that mean that the International standard becomes the United States national standard? So far it appears that our present trade standards are more stringent than the Codex standards. When white sugar begins to come from other countries, does that mean that we will reduce our quality in order to get down to their standards?

J. A. Hupfer: To answer your first question: yes, the Codex standard for white sugar would become our national standard to the extent to which the terms of the Codex standard are included within a FDA standard of identity or are encompassed within the basic requirements of the Federal Food, Drug and Cosmetic Act. As matter of practice, the U.S. government would have to promulgate a standard of identity based on a recommended Codex standard before formal acceptance could be given to the Codex standard.

For example, the recommended Codex standards for 5 sugars (glucose sirup), dried glucose sirup, dextrose monohydrate, dextrose anhydrous and lactose) were put into effect as standards of identity in this country before our government formally accepted the Codex standards for these products. The standards of identity for these sugars are identical to the Codex standards in most respects. However, it is important to bear in mind that you need only be concerned with the requirements in the FDA standards of identity and the basic requirements in the Federal Food, Drug and Cosmetic Act. In other words, the Codex standards have no legal effect except to the extent to which provisions in those standards are included in the food laws and regulations of our country.

In answer to your second question: Codex standards are intended to protect the interests of consumers, particularly those in developing countries which do not have adequate food safety laws or the means to enforce them. With this in mind, the Codex Alimentarious Commission decided that consumer interests can best be served by preparing standards which would assure sound, wholesome products of minimal acceptable quality. However, there is nothing in Codex food standards which would interfere with the perogative of food manufacturers and processors to market products at or above the minimal level of quality set in the standards. By the same token, requirements in Codex standards would not in anyway affect the right of consumers and users to choose higher than minimal quality products.

K. H. Schoenrock (Amalgamated Sugar): I notice you establish standards and specifications for the various products. But have you standardized on the analytical methods as well? I noticed the ICUMSA method only for color; how about the other methods?

J. A. Hupfer: Yes, each of the Codex standards for sugars contain a section on methods of analysis which contains references to the international

referee methods to be used in determining compliance with requirements in the standards.

There has been some discussion about methods among the Codex Committee, ICUMSA, and Dr. Horwitz of the Association of Official Analytical Chemists (AOAC), whose methods are used by FDA. The differences in methods are not great, and I anticipate that they will be resolved and the methods will be standardized.

C. B. Broeg (SuCrest): The published Codex International Standards for sugar contain ICUMSA Methods. The problem with regard to methods arises in the United States. My last conversation with Dr. Horwitz indicated that he has contracted with independent laboratories for the evaluation of some analytical methods used in Codex standards. In any event, he will insist on collaborative testing before he will recommend to the Food and Drug Administration that the Codex standards be proposed for adoption as standards of identity. Discussions with Dr. Horwitz are continuing.

C. C. Chou (Amstar): The industry's recommended pol for white sugar standard is 99.8. What is the chance of this being accepted?

J. A. Hupfer: Your recommendation was accepted by the U.S. government. However, we were not successful in our efforts to persuade the Sugars Committee to establish a requirement of not less than 99.8° polarization for white sugar. A majority on the Committee preferred 99.7° minimum for polarization.

A recommendation for a 99.8° minimum for polarization was also included in our government's official comments on the standard to the Codex Commission, but that body declined to overrule the Committee on this issue.

The only chance we would have of getting this recommendation accepted would be through an amendment to the recommended standard. However, it is unlikely that the Commission would approve amending the standard at this time.

J. C. P. Chen: The ICUMSA and the U.S. National Committee of that organization may provide methods of analysis for our acceptance, but the standards have to be determined by the industry. The Cane Sugar Refining Research Project has for some years been focusing its attention on analytical methods and identification of impurities and the possibilities of removing them from sugar. My main objective in inviting Mr. Hupfer to come here to talk to us was that we might learn new limits, or anything new in these standards about what we must remove from sugar so that we could go into any necessary research project on these matters. I think that it is very good to hear about what he is proposing even though the U.S. has not yet accepted those standards.

I think the S.I.T. should have a special committee or task force to work closely with the FDA, and to propose National Standards for final adoption.

NEW APPROACHES TO THE APPLICATION OF ION EXCHANGE IN THE SUGAR INDUSTRY

By Karlheinz W. R. Schoenrock, Avinash Gupta, and Dennis Costesso[1]

(Presented by Karlheinz W. R. Schoenrock)

ABSTRACT

Ion exchange was applied to eliminate selectively the noxious substances from sugar solutions. This new approach reduces waste handling and the cost of ion exchange significantly while still achieving the benefits of increased extraction and product quality. Techniques were also developed to convert all waste solutions into valuable byproducts. The primary impurities considered here are limesalts, color, floc and cationic impurities which inhibit the crystallization of sucrose.

INTRODUCTION

The basic processing steps for the manufacture of refined sugar from sugarcane or sugar beets have been essentially unchanged over the past 100 years. Only two major process innovations come to mind that represent really new technologies: Ion exchange and juice extraction via diffuser.

Ion exchange, despite its virtues in eliminating impurities not accessible by other means, has not been universally accepted in the sugar refining industry.

In reviewing the long-term survival of certain ion exchange systems in the refining of sugar, it becomes evident that it is in the areas of specific impurity elimination where success has been maintained. Attention is called to such established applications as softening and magnesium exchange via the Quentin process[2]. Hence, it appears that benefits from ion exchange are optimized when used to eliminate specific noxious impurities.

In this type of process, the main disadvantages of ion exchange are minimized while benefits are maximized. It is hardly a coincidence that the cost of regenerants is relatively low while the disposal of the waste does not generate major problems with these two ion exchange processes. It is, furthermore, evident that the application of magnesium exchange via Quentin is restricted to areas where an inexpensive source of magnesium chloride brine is available.

Technologies have recently been developed to advance the utility of ion exchange along this line in the processing of sugar beets as well as cane.

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[2]McGinnis, R. A. 1971. Beet-sugar technology. 2nd ed. pp. 330-333. Beet Sugar Development Foundation, Fort Collins, CO 80521.

Although all of the studies reported here have been directed toward the processing of sugar beets, there are certain phases which promise substantial utility in the processing of cane.

RESULTS AND DISCUSSION

Softening

The softening of solutions, that is the removal of calcium and magnesium, is universally practiced by operating over the sodium form of a strong cation exchanger and regenerating with common salt. Although salt is the lowest cost chemical available nearly everywhere, a large excess is required to reach a reasonable compromise between effective operating capacity of the exchanger, regenerant utilization, and waste disposal.

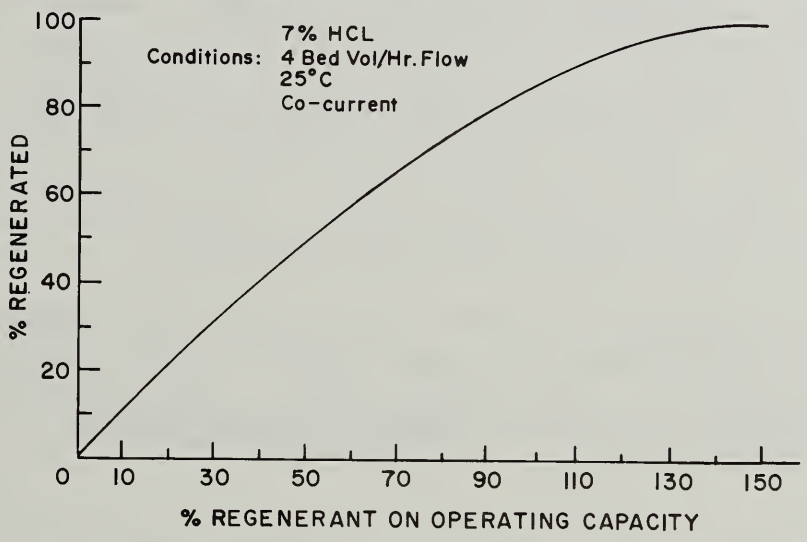


FIGURE 1--Regeneration profile for weak catex.

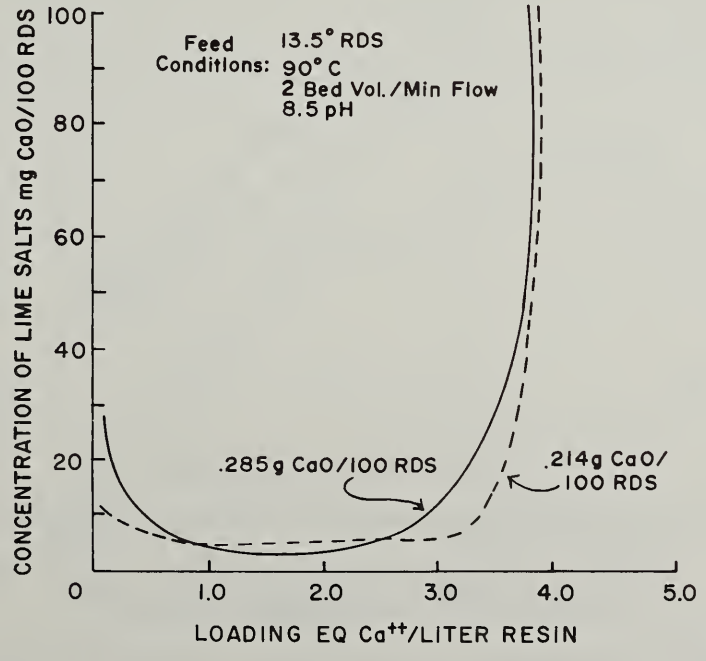


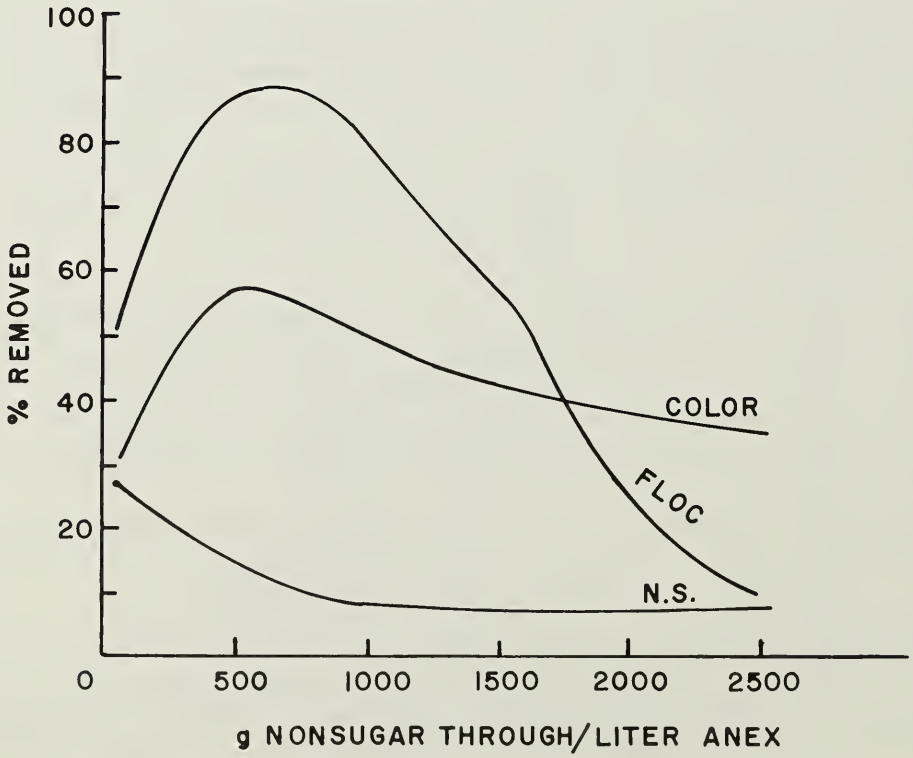
FIGURE 2--Ca^{++} leakage through weakly acidic catex.

It is generally known that weakly acidic cation exchangers have a great selectivity toward divalent cations yet can be regenerated with nearly stoichiometric amounts of acid. Figure 1 demonstrates a 90% conversion to the acid form with only 110% hydrochloric acid regenerant on the actual operating capacity of a weak catex (cation exchange resin).

A conventional type softener, operating over the sodium cycle of a strong catex, generates an operating capacity of only about 1 equivalent/liter exchanger with nearly 300% regenerant (salt) on operating capacity. A weak catex, operated over the hydrogen cycle, can yield operating capacities up to 3.5 equivalents/liter catex with only 110% regenerant on operating capacity. Figure 2 illustrates a typical loading cycle for a weak catex operated as a softener.

Waste disposal with the weak catex is, consequently, reduced to nearly one third of the load generated with the conventional softener process, while plant size is reduced to less than one third of that of the conventional softener. Another advantage is that the slightly acidic calcium chloride solution from the weak catex regeneration is ideally suited as an additive to diffuser operation to improve pulp pressability and raw juice quality. This waste solution is also far more effective in stripping and regenerating a strongly basic decolorizer to the chloride form than is the commonly used salt solution.

In separate pilot studies, this acidic $CaCl_2$ waste was, furthermore, very effective in stripping an exhausted weak anex (anion exchange resin) prior to its regeneration with ammonia to the hydroxyl cycle. The amount of ammonia required to maintain optimum operating capacity was thus reduced.



FICURE 3--Profiles for specific nonsugar after OH exchange on IRA 68.

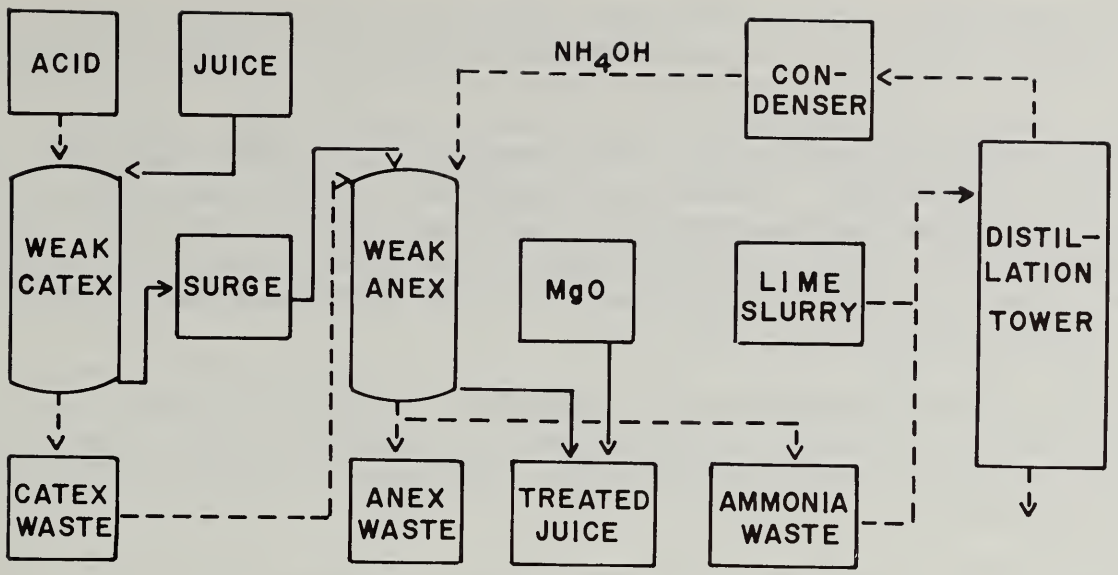


FIGURE 4--Weak catex - anex ion exchange flow diagram.

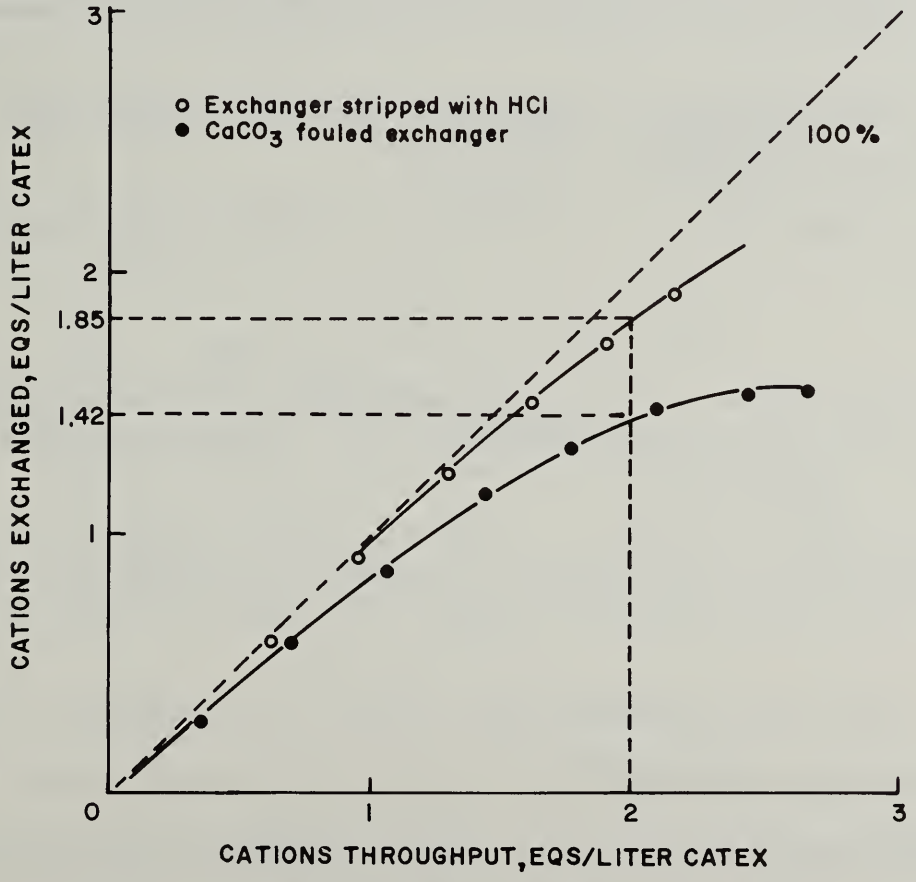


FIGURE 5--Loss in operating capacity due to $CaCO_3$ fouling.

In the softening of impure sugar solutions over the hydrogen form of a weak catex, the treated effluent is slightly acidic and must be realkalized to prevent sucrose inversion during evaporation and crystallization. A mildly acidic impure sugar solution will sufficiently activate a weakly basic exchanger in the hydroxyl form to restore most, if not all, of the required alkalinity required for further juice processing. At the same time, most of the floc and color will be eliminated while removing significant quantities of the impurities. Figure 3 shows a typical profile across the weak anex in the hydroxyl form. The ammonia used for the regeneration of the anex can be easily recovered from the regeneration waste through distillation with lime.

Additional active magnesium oxide, which has a very low melassigenic factor, may be added to a sugar solution treated by this process to assure sufficient alkalinity for sugar end processing. The entire process is graphically illustrated in figure 4.

The primary advantages of such a system are as follows:

1. Small plant size. 1 cu ft of a suitable weak catex can remove over 10 lb of calcium carbonate without re-introducing the highly melassigenic sodium ion.

2. Increased extraction over the conventional softening process. Nearly 15 lb of sugar are gained in extraction for each pound of sodium kept out of the sugar stream.

3. The waste load generated is relatively small since essentially stoichiometric amounts of regenerants are applied.

4. Some of the regenerants can be easily and relatively inexpensively recovered.

Total Cation Exchange

Another attractive approach is the exchange of the highly melassigenic metal cations for weakly melassigenic magnesium ion. This is practiced in the Quentin process but is limited there to an exchange of less than 50%.

Total cation exchange is, however, possible over the ammonium form of a cation exchanger followed by displacement of the exchanged ammonium in the juice with magnesium ion from active magnesium oxide. The reaction mechanism involved is shown below.

$$(\text{Catex}—\text{NH}_4) + (\text{Cat}—\text{An}) \rightarrow (\text{Catex}—\text{Cat}) + (\text{NH}_4—\text{An}) \quad \text{Exhaustion, (1)}$$

$$2(\text{NH}_4—\text{An}) + \text{MgO} + \text{Heat} \rightarrow (\text{Mg}—\text{An}_2) + \text{H}_2\text{O}\uparrow + 2\ \text{NH}_3\uparrow \quad \text{Realkalization, (2)}$$

$$2(\text{Catex-Cat}) + 2(\text{NH}_4)_2\text{CO}_3 \rightarrow 2(\text{Catex-NH}_4) + \text{Cat}_2\text{CO}_3/(\text{NH}_4)_2\text{CO}_3 \quad \text{Regeneration, (3)}$$

$$\text{and } \text{Cat}_2\text{CO}_3/(\text{NH}_4)_2\text{CO}_3 \rightarrow \text{Cat}_2\text{CO}_3 + (\text{NH}_4)_2\text{CO}_3\uparrow \quad \text{Regenerant Recovery, (4)}$$

where An = anion and Cat = cation.

The inherent advantages are, again, based on the low consumption of chemicals, the high effective exchanger utilization, and the associated low waste production.

The exchanger is first regenerated with a 40% $(NH_4)_2CO_3$ solution at a level of 200% on the total capacity of the catex, as shown in equation (3). Operating capacities of up to 1.8 equivalents/liter of catex can be realized if the impure sugar juice to be treated is free of calcium ion. Overall cation leakage is less than 10%. Figure 5 shows such an exhaustion profile.

The presence of calcium in the feed could foul the exchanger with precipitated calcium carbonate when regenerating with ammonium carbonate. A loss in operating capacity for subsequent cycles is the consequence of such fouling, as can be seen from figure 5. However, techniques have been developed which avoid this fouling, even when treating solutions containing high levels of lime salts.

Regeneration with ammonium carbonate allows recovery of excess regenerant by simple distillation. Ammonium carbonate volatilizes at about 56°C and can be recovered in the condensate as a 40% solution.

The carbonates of potassium and sodium remain in the bottom of the distillation column from where they can be recovered in fairly pure form. Pilot studies have demonstrated that this process could be operated without any waste discharge except for the backwash waters which carry a very low pollution load.

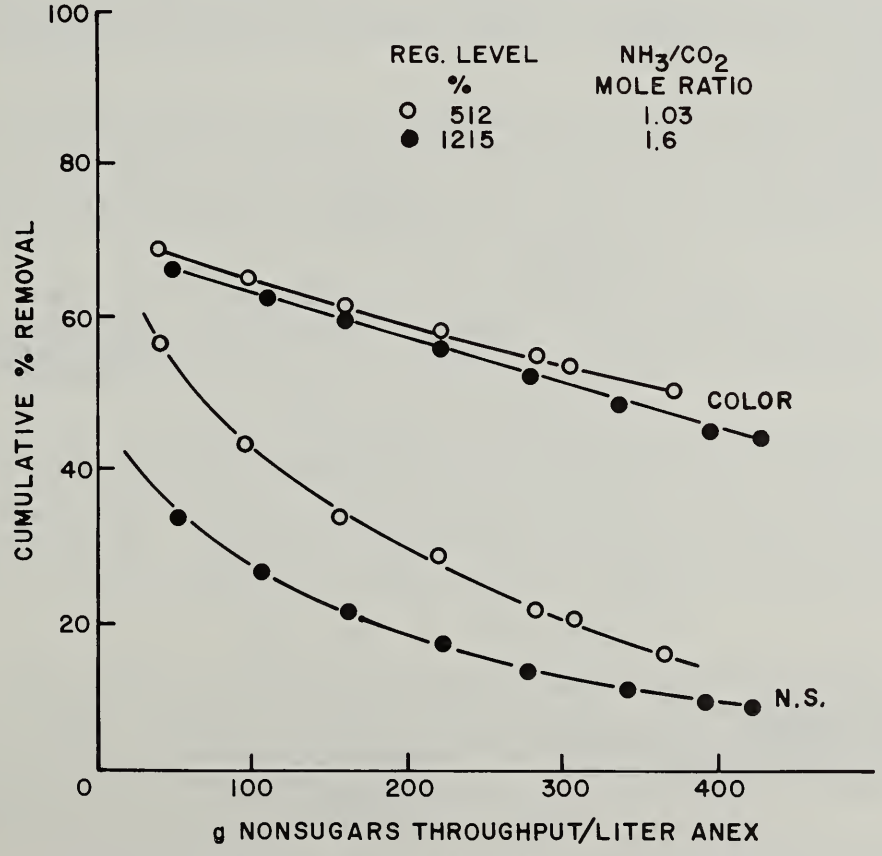


FIGURE 6--Effect of regenerant composition on anex performance.

The ammonia is displaced from the treated juice through the addition of active MgO according to equation (2) and is recovered from the vapor of the juice evaporator. Total makeup of ammonia is less than 10%.

Anion Exchange

A natural adjunct to this full cation exchange system is the operation of an anex in the carbonate/bicarbonate form. The bicarbonate form of a strong anex was found to operate substanitally more efficiently than its equivalent carbonate form. Figure 6 illustrates a comparison of impurity removal by both systems.

The removal of floc averaged around 90% in pilot studies. Including the anion exchange does not substantially increase the regenerant cost, but reduces the amount of active magnesium oxide required to displace exchanged ammonia. Figure 7 illustrates the recovery of exchanged ammonium ion and bicarbonate.

The reactions involved are:

$$\text{Juice} - NH_4HCO_3 + \text{heat} \rightarrow \text{juice} - NH_4 + \uparrow NH_3 + \uparrow CO_2$$

$$\text{Juice} - NH_4 + MgO + \text{heat} \rightarrow \text{juice} - Mg + \uparrow NH_3.$$

It is evident that all free ammonium carbonate must be expelled before the active magnesium oxide is added, to avoid formation of magnesium carbonate.

Figure 8 shows a flow diagram for the complete process which can be erected or operated in stages.

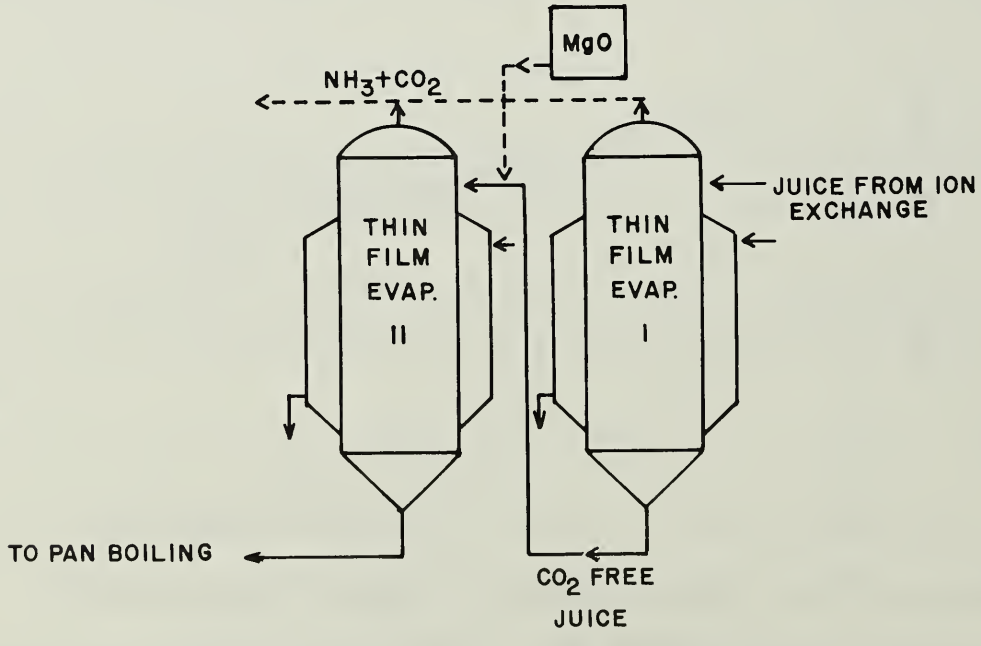


FIGURE 7--Regenerant recovery and MgO Neutralization of ion exchange treated juice.

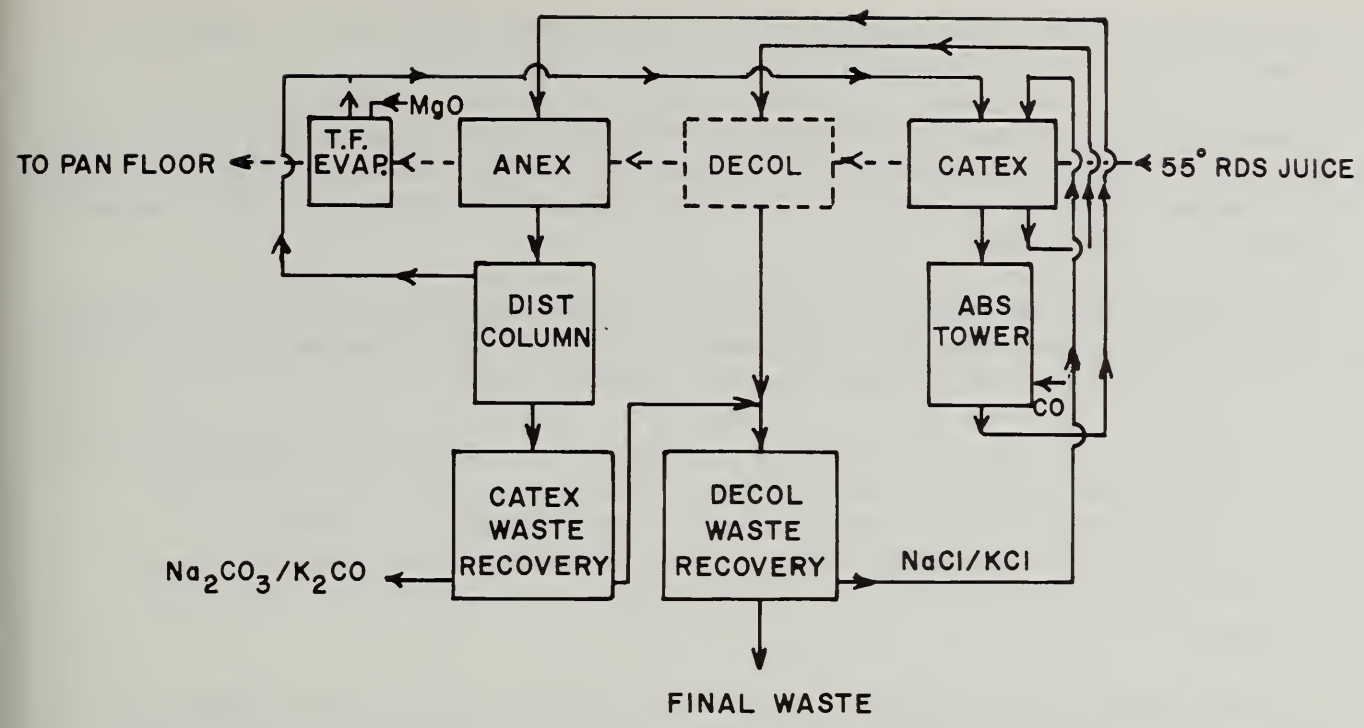


FIGURE 8--Outline of proposed ion exchange scheme
(Patent Pending)

A decolorizer operated in the chloride form and regenerated with internally recovered chloride salts may be added to protect the anex operated in the bicarbonate form from excessive fouling. According to figure 8, the complete process operates as follows:

1. If the impure sugar juice contains more than 0.02% CaO per 100 g dry solids (D.S.), it is taken over a two-stage catex in the ammonium form. The first stage will retain the calcium plus some potassium and sodium. The second stage will retain the residual juice cations until it is nearly loaded.

2. The juice, now containing only ammonium cations, may be taken through a decolorizer operated in the chloride form, and then through an anex operated in the bicarbonate form, or directly to the bicarbonate exchanger.

3. A thin film evaporator expells all free ammonium carbonate and bicarbonate from the treated juice coming out of the bicarbonate exchanger. Active magnesium oxide is added to the juice discharged from No. 1 evaporator that is now free of carbonates. A second evaporator serves to expel residual ammonia after addition of active magnesium oxide. After this, the juice is ready for pan boiling.

4. The first stage of the catex, which contains substantial amounts of calcium ion, is first stripped with a decalcified salt solution recovered from the decolorizer waste. This process removes the calcium ion. This is followed by the regenerating of both catex stages with a saturated solution of ammonium carbonate recovered from the distillation tower.

5. The calcium stripping solution from the first stage catex is used to regenerate and strip the decolorizer.

6. The cation regeneration effluent, containing excess ammonium carbonate and all exchanged juice cations in their respective carbonate form, is now treated with carbon dioxide to convert the carbonates to bicarbonates that then serve to regenerate the anex.

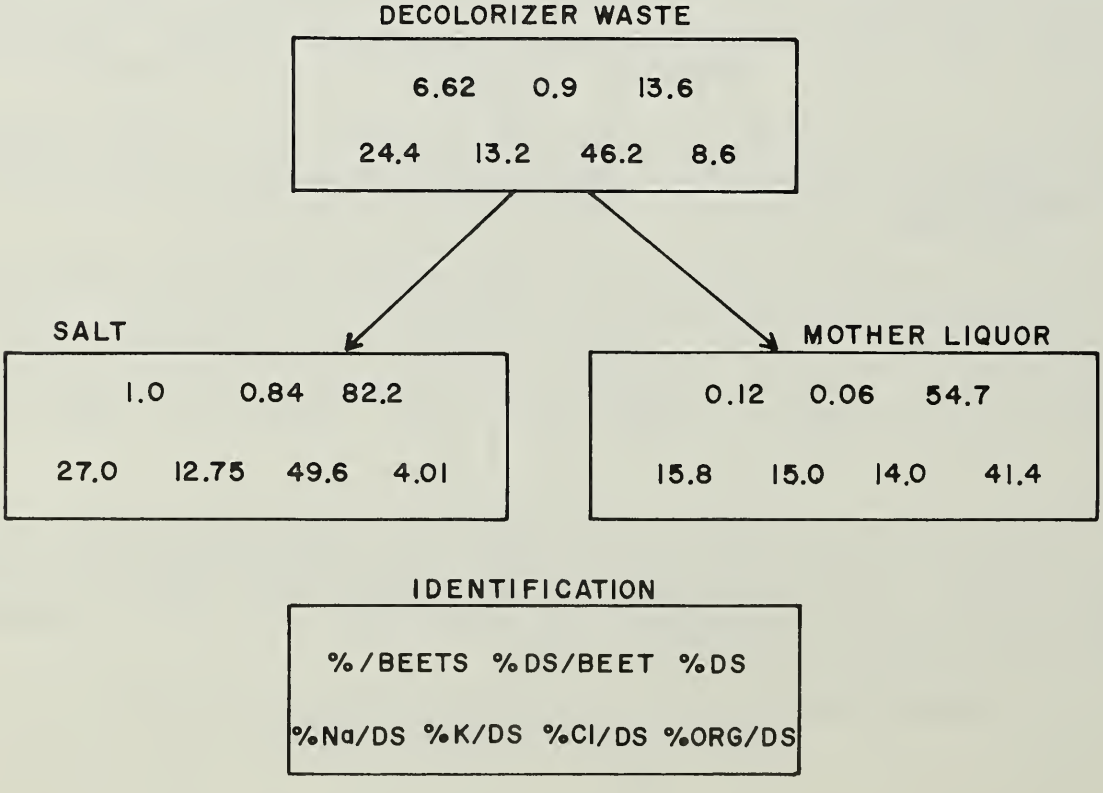
7. A distillation column operated by the ammonia-bearing vapors from the evaporators recovers excess ammonium carbonate which, in turn, is used to regenerate the catex to the ammonium form as outlined above, under No. 4.

8. The bottoms from the distillation tower which contain predominantly potassium and sodium carbonate are treated by evaporative crystallization to recover these carbonates in their respective crystalline forms. The final mother liquor, which contains about 10% of the alkali metal carbonates, is used to decalcify the decolorizer waste by precipitating the calcium as insoluble calcium carbonate.

9. Potassium and sodium chloride are recovered from this decalcified waste through evaporative crystallization and light calcination at 400°C. The salts are then used as a calcium stripper solution in the first-stage catex.

The primary advantage of this ion exchange process lies in the internal recovery of required regeneration chemicals, and the practical elimination of waste pollutants. The operating costs will thus be kept low.

TABLE 1--Decolorizer waste basic composition



According to the scheme outlined in figure 8, only one final waste solution goes out of the system. It is a product high in organic nutrients; hence, suitable as a cattle feed. Pilot studies have established values for a meterial balance around the salt recovery as shown in table 1.

Accordingly, the final waste from this operation represents only 0.12% weight on the beets processed and is already in a very concentrated form: 54.7% D.S. and over 40% organic matter on dry substance. More than 90% of the salt was actually recovered through simple evaporative crystallization. The most efficient crystallization was found to be a three-stage operation without recycle, as shown in table 2.

TABLE 2--Economic analysis of common salt recovery from decolorizer ion exchange waste

Set no.	% D.S., original stock sol'n.	Yield, % crystallization				Purity % ash D.S.	Cost of steam T.D.S., at $1-5/10^6 BTU	Value of recovered salt at $20/T.D.S.
		A stage	B stage	C stage	Overall			
1	14.0	50.2	66.9		83.5	96	7.17	16.70
2	14.0	47.7	68.8		83.7	97	6.54	16.74
3	14.0	33.1	61.6	72.6	93.0	96	6.80	18.60

The final salt product, containing about 4% organic matter carryover, was slightly calcined and yielded an excellent salt for softener and decolorizer stripping, since it is essentially free of calcium.

The recovery of the potassium and sodium carbonates from the distillation bottoms, as projected in figure 8, is carried out very similarly. Table 3 shows the basic values for the feed, product, and mother liquor. Again, the total quantities as percent on beets are fairly small, at around 0.5% beets. Over 90% of the exchanged juice cations are recovered as carbonates from the distillation bottoms. The three-stage crystallization gave, again, the best efficiency as shown in table 4. Part of this improvement was achieved by improved fractionation during regeneration to reduce the dilution and energy cost during subsequent concentration of the distillation bottoms.

As pointed out above, the final mother liquor from this soda ash/potash recovery is combined with the salt waste from the softener or decolorizer to precipitate all soluble calcium salts as calcium carbonate (as shown in figure 8), prior to the recovery of sodium chloride.

The large differences in the solubilities of sodium and potassium carbonates and their respective bicarbonates allow large flexibilities in the separation of the various salt components from each other. It is, therefore, possible

TABLE 3--Catex waste basic composition

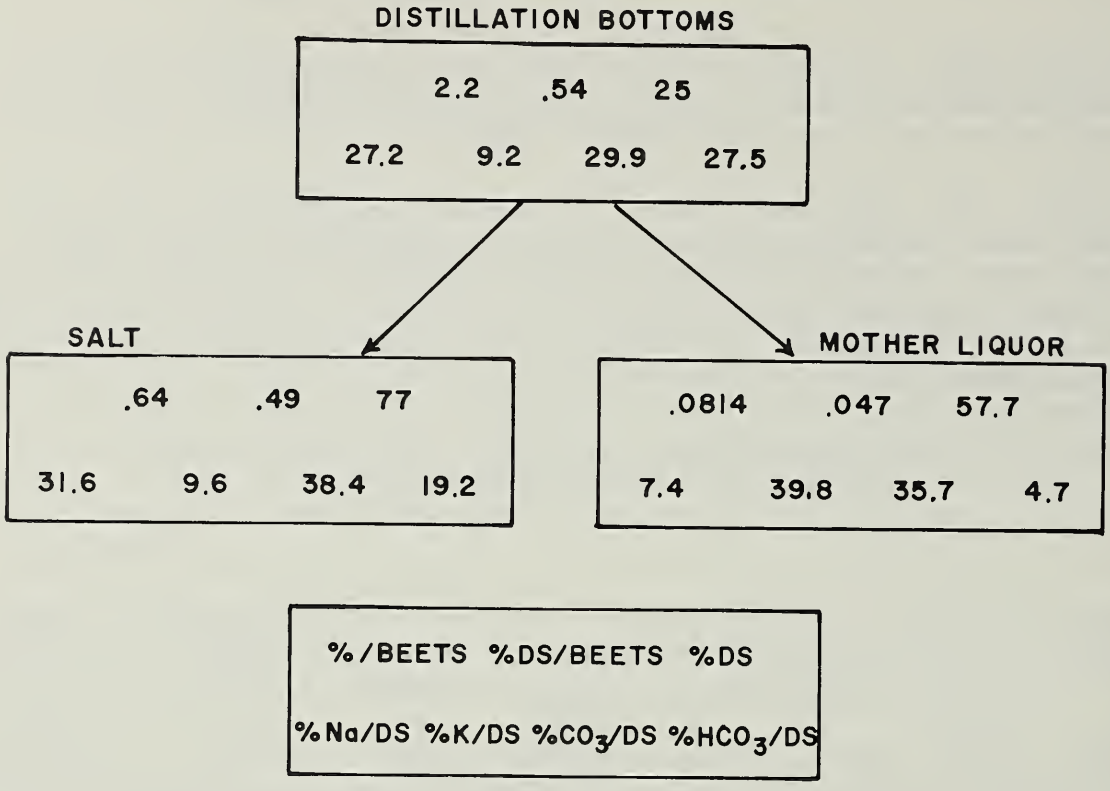


TABLE 4--Economic analysis of potash/soda ash recovery from catex ion exchange waste

Set no.	% D.S. Dist. bottoms	Yield, % crystallization				Color units/ 100 R.D.S., over-all	Cost of steam T.D.S., at $1-5/10^6$ BTU	Value of recovered salt at $30/T.D.S.
		A stage	B stage	C stage	Over-all			
1	14	42.3	69.3		82.3	252	12.42	24.69
2	14	74.3	65.7		91.2	337	15.15	27.36
3	25	35.0	35.7	79.3	91.3	293	7.47	27.39

to produce fractions which are either predominantly soda ash or potash, or mixtures of these two.

CONCLUSION

Every phase of these ion exchange systems has actually been carried out in pilot tests with varying potassium/sodium ratios. It was deomonstrated that the

various phases of the entire process system are relatively simple operating steps. Each operating step represents a well-proven and commercially successful unit operation.

The process can be erected in stages, where each stage will operate on its own merits, but can also provide the prerequisite for further refinements in the elimination of noxious impurities.

The process is also largely self-supporting with regard to the required regeneration chemicals. Only carbon dioxide needs to be furnished, either from lime kiln gasses or boiler stack gasses.

The problems of waste disposal commonly associated with ion exchange operations are effectively eliminated in this system of new approaches to the applications of ion exchange resins.

DISCUSSION

C. C. Chou (Amstar): What would be the capital cost for this system for a 2×10^6 lbs per day plant?

K. H. Schoenrock: Considering the first softening process, for a plant slicing 6,500 tons beets/day and removing 250 mg CaO per 100 g dissolved solids, it would be under a million dollars, capital cost. This is for about 1,000 tons sugar introduced at 16% sugar on beets.

C. C. Chou: In the recovery of ammonia, what would be the percent recovered?

K. H. Schoenrock: In both cases, it is over 90%. Ammonia is very expensive and getting more so.

W. L. Reed (Revere): What about the energy application? It seems like a very nice scheme in which you solve pollution problems, but does it take a great deal of energy? Are we reducing pollution but with a net increase in energy usage?

K. H. Schoenrock: It probably will utilize energy better than the present ion exchange system for two reasons. One, it is operated at the normal juice temperature. Conventional purification systems with strong cation exchange in the hydrogen form require cooling because of the danger of inverting the sugar if it were not cooled. Operation over the ammonium form does not require cooling and reheating, thus eliminating a large drain on the energy resources. In this latter case, the energy required for normal juice concentration is also utilized to recover the ammonia in the juice. Recovery of ammonia from the regenerant represents a smaller energy drain than that usually used any way in production.

W. L. Reed (Revere): In the recovery of all the reagents which you have described you probably still need a modest amount of makeup. Would you have an idea of the use of this? Is it negligible?

K. H. Schoenrock: Largest makeup requirements involve carbon dioxide. Ammonia makeup is estimated to be 5% to 10%. The juice would normally provide all the cations needed to sustain the process. A beet sugar factory slicing 10,000 tons of beets per day may generate 30 tons to 50 tons of potassium and sodium carbonate per day to sell.

R. Moroz (Sucrest): I gather that you were working at 50 Brix density?

K. H. Schoenrock: Between 50 and 60 R.D.S.

R. Moroz: If you have invert in the syrup, what is the relationship between invert and color? In a cane refinery there is substantially more invert than in a beet factory and if you used ammonia on cane liquors you would have a tremendous color problem. There would be color formation rather than color removal.

K. H. Schoenrock: That is part of the reason why we are using the thin film evaporator; with its very short contact time, it helps to eliminate the formation of color until after the ammonia has been replaced. After ammonia is eliminated there are no differences. Of course, we have not done this kind of study with cane sugar, but I visualize that the problem would not be significant because of the short contact time in the thin film evaporators. There are other ways of eliminating the problem of invert. I don't know whether the cane industry has thought of trying to destroy the invert and then to eliminate the breakdown products of the invert.

R. S. Patterson (C&H): You've indicated that this has been done on a pilot-plant scale. What are the plans to move to commercial?

K. H. Schoenrock: We're waiting for the sugar prices to come up.

THE ROLE OF COMPOSITION OF LIQUOR ASH AND OF THE IONIC FORM OF THE RESIN IN DECOLORIZATION OF REFINERY SIRUPS

By Milan Vender

ABSTRACT

Decolorization of refinery sirups by strong base resins is an ion exchange process in which the ionized colorants compete for resin sites with inorganic and low molecular weight organic anions contained in the sirup. Selective preferences of resin for different ions and colorants can significantly influence the decolorizing performance. It thus seems possible to improve the decolorizing process by using a suitable initial ionic form of the resin and by manipulating the composition and concentration of ash in the sirup prior to decolorization.

A two-stage process mechanism is suggested; in the first stage the resin is partly converted from the initial ionic form to the form which is in equilibrium with liquor ash. This form in turn participates in the exchange reaction with colorants. The model explains the observed phenomena.

INTRODUCTION

For a long time the mechanism of liquor decolorization by ion exchange resins was quite obscure. Published information[1,17,23] indicated both ion exchange and adsorption as the principal processes. Experimental findings at CSR Laboratories seemed incompatible within themselves and were difficult to explain: whereas some experiments indicated a straightforward relationship between the ion exchange capacity of the resin and ability to decolorize, others suggested that such a relationship is almost nonexistent. While some raw liquors were easily decolorized, others - for no apparent reason - resisted decolorization. Sophisticated methods were used in attempts to separate and identify troublemakers among the colorants but produced no significant results.

When the application of ion exchange resins to decolorization of refinery liquors was studied, it was soon discovered that the key to the understanding of the decolorizing mechanism lies in the mutual interactions of resin with colorants as well as with inorganic and low molecular weight organic electrolytes.

In this paper are reported some of the most important findings from this study, which contribute to the understanding of the decolorization process.

EXPERIMENTAL

A block diagram of the experimental column together with normal working conditions is shown in figure 1. The assembly is automated and works round the

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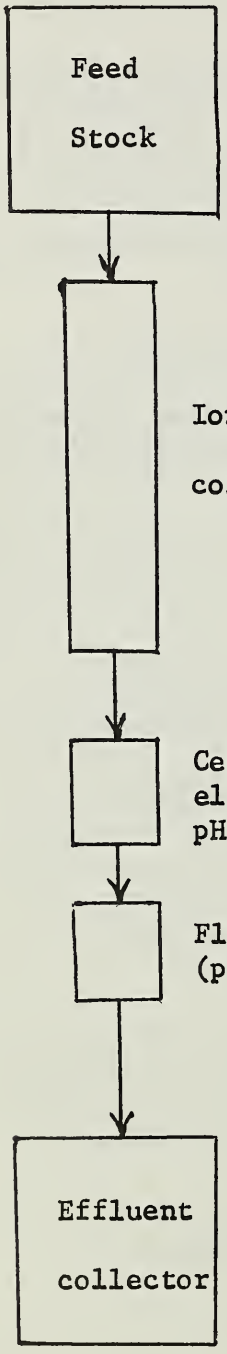
clock with minimum attention. Monitoring of effluent parameters can be easily modified or extended; for instance, the concentration of selected ions could be monitored using ion selective electrodes. The optical absorbance of the effluent could also be monitored on line, but more accurate methods with correct adjustment of pH were preferred.



Conditions:

Temperature	75.0 ± 0.5°C
Flow rate	3.0 ± 0.1 BV/h
Normal cycle	60 BV of feed 3 BV of regenerant
Feed	60-65° BX
Color (420nm, pH9)	1-2 x 10^6 CU m^{-3}
pH	8-9
Regenerant	100g/l NaCl

pH adjusted by caustic soda to 12.5

Resin: strong base anion exchange

resin, isoporous polystyrene matrix crosslinked by divinylbenzene, trimethylbenzyl-ammonium groups.
Volume of the bed 5 - 15 cm^3

FIGURE 1--Block diagram of experimental equipment.
Experimental conditions.

Precise analyses of feed and effluent are essential for the assessment of material balances of the operation. Most of the ions studied were determined by fully automated potentiometric and conductimetric titrations which gave the desired accuracy.

The concentration of colorants was measured as absorbance of light at a wave length of 420nm, in a 1 cm cell against distilled water, in filtered samples adjusted separately to pH 4.00 ± 0.05 and 9.00 ± 0.05 (15). A unit of the quantity of colorants (CU) is defined thus: 1 cm^3 of the solution contains 1 CU when the absorbance equals 1.00. The indicator value, which is the ratio of absorbances measured at pH 9 and 4, was used to represent relative proportions of pH-sensitive and pH-insensitive colorants in a mixture (18).

Sometimes it was necessary to characterize colorants more specifically. In such cases, UV and visible spectra, gel filtration, thin layer chromatography and electrophoresis were used for separation and identification of various classes of colorants and individual colorants (9,10,18-22,25).

The ionic form of the resin, its moisture content, the specific volume and porosity of the bed, together with pH titration curves, were used to characterize the chemical, hydrodynamic and ion exchange properties of the resins tested.

The evaluation of each experiment was based on material balances of sugar solids, color-units and equivalents of counter-ions.

MECHANISM OF DECOLORIZATION

Several studies have indicated that color is a mixture containing weak acids and bases (1,2,4,5,6). The well-known indicator effect is based on the different specific absorbances of the ionized and the undissociated forms of colorants. Electrophoretic separations reveal the presence of both cationic and anionic species in the colored mixtures. Practically all colorants contain various functional groups that are able to dissociate when the pH of the solution is sufficiently high or low.

The amphoteric nature of mixtures of colorants is shown by ample experimental evidence. For example, figure 2 is a typical plot of titration curves of resin used in decolorizing refinery liquors. The first curve was obtained with the resin fully converted to the hydroxide form. In this form, it behaves like a mixture of a strong and weak base. The same resin, after extensive regeneration with an alkaline solution of sodium chloride, yielded titration curve No. 2. This is the curve of a resin fully converted to the chloride form that behaves like a neutral salt. Finally, curve No. 3 shows the same resin loaded with colorants from raw liquor. It is apparent that this resin can neutralize not only strong acid but also strong caustic, as would be expected from an ampholyte.

Figure 3 shows similar behavior observed in resin during regeneration. In this diagram, the quantity of colorants extracted from a given amount or resin is plotted against the pH of regenerants used. In all cases a constant volume of 1 N sodium chloride solution was used as regenerant, with the pH adjusted by hydrochloric acid or sodium hydroxide. Absorbances were measured with pH

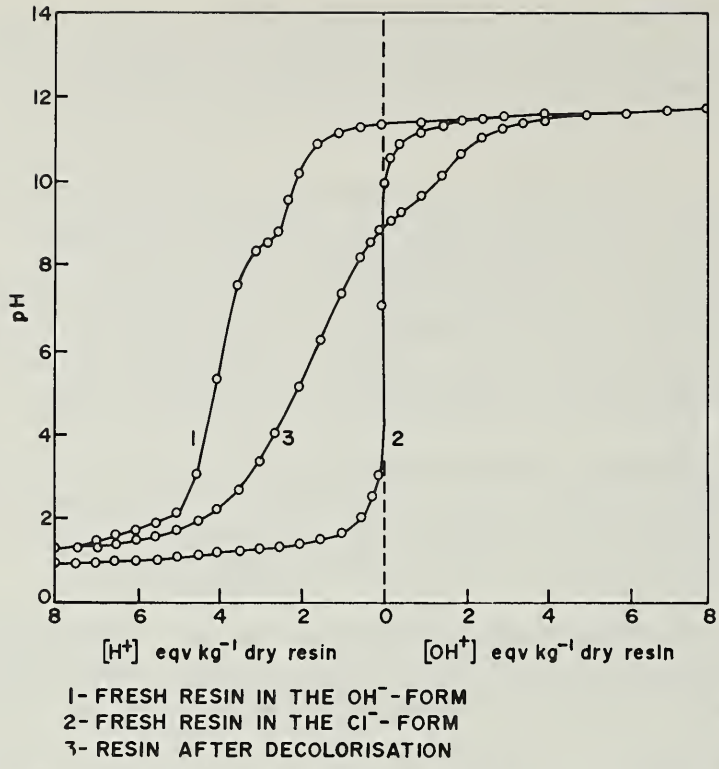


FIGURE 2--A typical plot of titration curves of decolorizing resin.

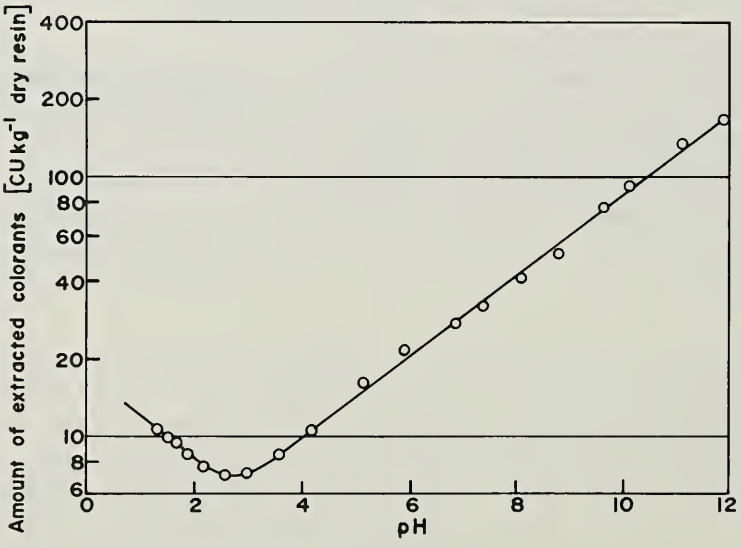


FIGURE 3--Effect of the pH of regenerant on the release of colorants from the resin.

adjusted to 9.00 ± 0.05. It is interesting to note that the pH at the lowest efficiency of regeneration is within the range of isoelectric points for some typical colorants.

The symmetrical shape of the curve seems to suggest that both strong base anion exchangers and strong acid cation exchangers could decolorize efficiently. However, in practice, only strong base exchangers are suitable because of infusion problems with strong acid exchangers. The best decolorizing effect is achieved when the pH is sufficiently high to dissociate colorants, but not high enough for hydroxide ions and even ionized sucrose to compete with colorants for the ion exchange sites of the resin.

Sugar colorants have been found to have electrochemical properties which make them suitable for ion exchange. Direct evidence that such exchange takes place is given by changes in electrical conductivity and in the concentration of anions in liquor during decolorization. Data from tests of different resin samples decolorizing the same feed are shown in table 1.

The specific conductivity, which is approximately proportional to the content of electrolytes in the solution, increased by about 27% over the initial value in the feed. By comparing the equivalent conductances of the most common inorganic and organic anions present in refinery liquors with that of chloride ions we find that the exchange of those ions for chloride could not change the conductivity by more than 5%. On the other hand, colorants have relatively large ionic radii and hence low equivalent conductances. If, in decolorization, the low-conducting colored ions in liquor are replaced by mobile chloride ions, the specific conductivity of the liquor must increase significantly. This increase was actually observed.

The last two columns of table 1 show the ratio of the quantities of colorants and anions exchanged. Bearing in mind that this measurement of the quantity of colorants is also influenced by the composition of the colored mixture, the variation of the ratio is within acceptable limits. The mean values of the ratio can be used to estimate resin decolorizing capacity.

In an attempt to measure the decolorizing capacity of the resin, a breakthrough curve, of colorants over 1000 bed volumes of liquor, was measured. After this volume of liquor was passed, the mean decolorizing efficiency was about 40% and the resin still had some decolorizing potential. With the figures from table 1, and the ion exchange capacity of the particular batch of resin at 0.8 keqv m^{-3}, the decolorizing capacity of 1 m^3 of resin bed was about 1.6×10^8 CU at pH 4, or 4.4×10^8 CU at pH 9. Thus, with a typical color concentration in feed of 0.36×10^6 CU m^{-3} at pH 4, or 1.0×10^6 CU m^{-3} at pH 9, a particular sample of resin would decolorize about 1100 bed volumes of liquor before being completely saturated. This calculation agrees with the direct measurement of the breakthrough curve.

This experimental evidence appears sufficient to illustrate the ion exchange mechanism of decolorization in the case of the strong base resin and liquor prepared from Australian raw sugar.

THE COMPETITION BETWEEN COLORANTS AND ELECTROLYTES

Liquors contain, in addition to colorants, inorganic and low molecular weight organic electrolytes which can also participate in ion exchange. These

TABLE 1--Ion exchange in decolorization of refinery liquors

Resin sample	Cycle no.	Increase of specific conductivity of liquor, $\times 10^{5}$ ohm^{-1} cm^{-1}	Anions exchanged, meqv	Amount of colorants exchanged (CU)		Colorants exchanged per anions exchanged, (CU)/meqv	
				pH 4	pH 9	pH 4	pH 9
1	1	5.8 ± 0.8	0.87 ± 0.12	277	791	318 ± 46	909 ± 127
	2	6.8 ± 0.9	1.32 ± 0.17	270	748	204 ± 28	566 ± 96
	3	6.6 ± 0.9	1.23 ± 0.17	269	746	218 ± 37	606 ± 103
2	1	7.1 ± 0.9	1.46 ± 0.19	277	771	189 ± 24	578 ± 67
	2	5.5 ± 0.9	0.74 ± 0.12	269	747	364 ± 60	1010 ± 165
	3	6.3 ± 0.9	1.10 ± 0.16	266	730	242 ± 35	665 ± 95
3	1	7.1 ± 0.9	1.46 ± 0.19	278	808	191 ± 25	553 ± 72
	2	7.3 ± 0.9	1.55 ± 0.19	271	764	175 ± 22	493 ± 61
	3	7.2 ± 0.9	1.51 ± 0.19	265	725	176 ± 22	482 ± 60
4	1	7.0 ± 0.9	1.41 ± 0.18	276	783	195 ± 35	553 ± 100
	2	5.8 ± 0.9	0.87 ± 0.14	272	775	312 ± 44	891 ± 125
	3	6.7 ± 0.9	1.28 ± 0.17	262	737	205 ± 27	576 ± 75
5	1	6.8 ± 0.9	1.32 ± 0.17	275	792	208 ± 28	600 ± 79
Mean	-	6.6 ± 0.6	1.24 ± 0.27	-	-	200 ± 20	560 ± 50

substances are natural competitors of colorants. Depending on their quantity and composition they may affect the process and influence the decolorizing performance (13). Work in this laboratory has been able to establish that in most cases the difference between easily decolorized and decolorization-resistent liquors was due more to the ash than to peculiarities in the composition of colorants.

Theoretically, a comprehensive description of the decolorization operation could be achieved by a rigorous thermodynamic and kinetic analysis of the process (8,11). In practice the necessary data to do so are not available. The possibility of ever being able to measure concentrations of all substances present in a particular liquor, evaluate their activities and know the values of their diffusivities is very small. These are the data needed for a description of equilibrium conditions, calculation of mass transfer rates and mathematical simulation of a fixed-bed decolorizing column.

On the other hand much useful information can be deduced by a less sophisticated application of basic thermodynamic and kinetic principles in combination with an experimental study of the roles of some key components. This was the approach which was used in this work.

There are three groups of reversible reactions which can simultaneously proceed in the resin - liquor system, namely:

$$(R^+ - Cl^-) + Col_i^- \rightleftarrows (R^+ - Col_i^-) + Cl^-$$

$$(R^+ - Cl^-) + El_j^- \rightleftarrows (R^+ - El_j^-) + Cl^-$$

$$(R^+ - El_j^-) + Col_i^- \rightleftarrows (R^+ - Col_i^-) + El_j^-$$

where $i = 1,2,---,$
$j = 1,2,---,$

and R^+ indicates the resin matrix with active sites,

Col_i^- represents a particular ionized colorant,

and El_j^- represents a particular non-colored anion in the liquor.

The first reaction is the exchange of chloride ions for colorants. The second is the competing exchange of non-colored anions from liquor ash and third is the decolorzing reaction with the resin converted to the ash-anionic form.

The amount of color removed by the first and the amount by the third reaction depend on the equilibrium and kinetic conditions. Some indication of the equilibrium is given by the sequence of preferences of the resin for particular ions (selectivity sequence). In the case of strong base resin the usual sequence of common ions is:

$$(PO_4^{3-}) > (SiO_3^{2-}) > SO_4^{2-} > NO_3^- > Cl^- > (CO_3^{2-}) > OH^- \quad (11,16).$$

Anions in brackets are included in positions indicated by their influence on decolorization in these experiments.

For a given colorant, better decolorization can be expected when the colorant substitutes for chloride than for, say, sulfate ions. In the competition between colorants and electrolytes, sulfate ions are more serious competitors than hydroxide, chloride, or nitrate ions.

The most important implication of the selectivity sequence is that the ionic form of the resin can significantly influence the decolorizing performance (16). This is demonstrated in figure 4 which shows the decolorizing efficiency of various ionic forms of the resin as a function of the mean concentration of colorants on the resin. These experimental data confirm expectations of ionic behavior.

The effective ionic form of the resin depends not only on the initial form but also on interactions with liquor ash and on the impurities in water and in regenerant solutions. It was found, for instance, that using a sand bed for filtration of alkaline regenerant solution can lead to a serious contamination of the resin by silicate ions. Occasionally, up to 56% of the total capacity was converted to the silicate form, and only repeated washing with hydrochloric acid could help to reduce the silicate contamination.

In continuous fixed bed ion exchange, the rate of transport of exchanged ions from the bulk of the liquor to the resin surface, and through the resin matrix to the active sites, and vice versa, is even more important than thermodynamic equilibria.

Fast-diffusing ions can accumulate in the vicinity of active sites in sufficiently high concentrations to counterbalance possible adverse equilibrium

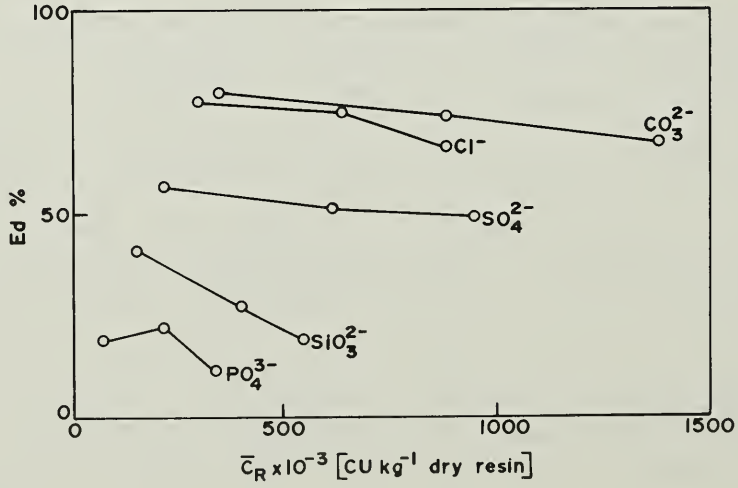


FIGURE 4--Effect of the initial ionic form of the resin on decolorization.

E_d = efficiency of decolorization

C_r = average color concentration (pH 9) on the resin over the cycle

conditions. Thus, under dynamic column conditions, the selectivity sequence is not necessarily followed and fast ions are preferentially exchanged even if the equilibrium favors slower ions. There is, however, a continuous change of these preferences, and as the process continues the slower ions replace the faster ones bringing the whole system to a state of thermodynamic equilibrium between the resin and feed liquor.

In order to describe the dynamic behavior of the ion exchange column in a more quantitative manner we have prepared a simplified scheme which is presented in figure 5. The scheme is developed from the following assumptions:

1. There are two kinds of competitive ions in the feed - fast (El_f^-), and slow (El_s^-) ions. Inorganic and small organic anions may represent the first kind and colorants the second one.

2. The affinity (equilibrium concentration ratio) of slow ions towards the resin is higher than that of fast ions. (In the given example, 10 times higher.)

3. The adsorption isotherm is linear (equilibrium concentration ratio is constant).

4. Mass transfer from the bulk of the liquor to the surface of the resin controls the overall rate of the process. The mass transfer coefficient of the fast ions is larger than that of slow ions. (In the given example, 100 times larger.)

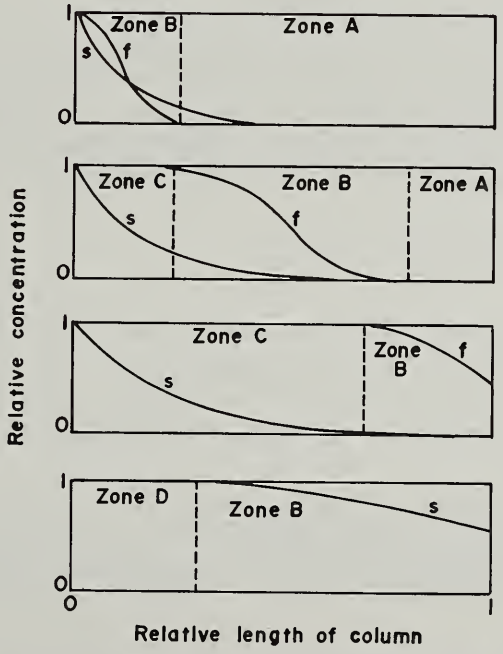


FIGURE 5--Schematic presentation of the dynamics of the ion exchange column.

s - breakthrough curve of slow ions (colorants)

f - breakthrough curve of fast ions (ash anions)

5. The differences in selectivity between the slow and fast ions on the one hand and the slow and initial resin ions (El^-_r) on the other are small.

With these assumptions, Schumann's model of the fixed bed process can be used for the construction of breakthrough curves within the bed[3]. We can see that the breakthrough curve of fast ions divides the bed into several zones that move slowly in the direction of the flow.

At the start of the operation, zone A is spread over the whole bed and all resin is in the initial ionic form ($R^+-El^-_r$). However, immediately after the introduction of feed into the column, a second zone, B, is formed. In this zone the resin is already partly converted to the ($R^+-El^-_f$) form by the exchange reaction with the fast ions:

$$(R^+-El^-_f) + El^-_f \rightleftarrows (R^+-El^-_f) + El^-_r.$$

In zone B slow ions are exchanged both for initial resin ions (El^-_r) and fast ions (E^-_f) to a degree which depends on the relative concentration of both forms. This can easily be found from the breakthrough curve.

Generally, in the region on the right side of the breakthrough curve for fast ions, the exchange reaction

$$(R^+-El^-_r) + El^-_s \rightleftarrows (R^+-El^-_s) + El^-_r$$

prevails, whereas on the left side of the curve, the main reaction is

$$(R^+-El^-_f) + El^-_s \rightleftarrows (R^+-El^-_s) + El^-_f.$$

With continued pumping of feed into the column, zone A is slowly reduced, zone B becomes wider, and a new region (zone C) appears. In this latter zone all available resin is converted to the ($R^+-El^-_f$) form and the main exchange reaction is between the fast and slow ions:

$$(R^+-El^-_f) + El^-_s \rightleftarrows (R^+-El^-_s) + El^-_f.$$

Finally, the complete conversion of the bed to the ($R^+-El^-_f$) form is accompanied by the disappearance of zone B. All exchange proceeds in zone C and a final new zone (D) appears. This is the zone which contains the resin in equilibrium with the feed, and in which no further exchange reactions proceed.

Earlier studies in this laboratory demonstrated that mixtures of colorants behaved in diffusion as a single component with the diffusion rate controlled by large molecules[24]. The diffusivities of common ions also lie in a relatively narrow range.

Thus, for instance, the numerical value of color diffusivity in a 65 Bx liquor at 70°C was 1.84×10^{-10} $m^2 s^{-1}$, as compared with 4.04×10^{-10} $m^2 s^{-1}$ for sucrose under the same conditions[12]. Electrolytes diffuse even faster than sucrose. Cussler and Dunlop[7] measured ternary diffusion coefficients for a

system containing water, sucrose and potassium chloride at 25°C at concentrations of 30 g of sugar and 60 g of potassium chloride per liter. They found that the diffusivity of KCl was 3.5 times that of sucrose. It is not possible to extrapolate these figures to higher sugar concentrations (i.e. to 850 g/l) but it is highly probable that electrolytes do diffuse faster than sugar in sugar liquors. We can therefore safely assume that, of the competing species, small colorless anions have the best chance of reaching the active sites of the resin first.

Under these conditions the model representing the competition of slow and fast ions seems to describe the decolorizing process as a good approximation. It can be assumed that, with the exception of the earliest stages, decolorization proceeds as a two-stage process: in the first stage the main exchange reaction is the substitution of chloride (or other initial ions) from the resin by anions introduced in the liquor ash. The second stage is the decolorization itself, in which colorants substitute for ash anions in the converted resin.

The ionic form of the resin also has an indirect effect on decolorization by influencing the pH of the liquor. When strong anions such as sulfates and chlorides are exchanged nothing much happens and the pH of the product is close to the pH of the feed (see figure 6). On the other hand carbonate, silicate, and phosphate forms of the resin have quite dramatic effects. These relatively weak anions tend to raise the pH of the product above that of the feed. In the cases of phosphate and, to a lesser degree, carbonate ions the effect of various neutralization steps is also apparent.

EFFECTS OF SOME CATIONS ON DECOLORIZATION

Attention was directed to calcium, magnesium, iron, and copper. These are the ions with high probability of occurrence that are able to produce adverse effects on the resin and on the quality of the decolorized product (14).

The effect of calcium and magnesium was studied from the aspect of possible contamination of the resin. Also, these ions are probable precursors of high

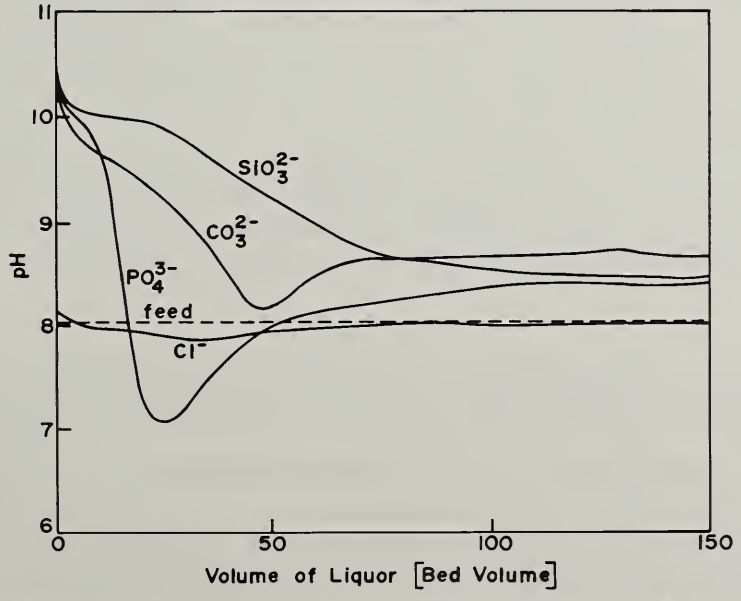


FIGURE 6--Effect of ionic form of the resin on the pH of product.

turbidity in decolorized liquor. A comparative study of resin washed with distilled water and regenerated by reagent purity chemicals with the same resin treated by normal factory tap water and technical grade chemicals indicated that, over ten cycles, the average color concentration of product was better by 17% in the case of pure water and chemicals. Calcium and magnesium salts were the main impurities in factory water and salt. The observed difference was statistically significant at a 99% probability level.

Analyses of service resin showed that, in most cases, the content of calcium and magnesium was less than 0.3% of the ion exchange capacity. This means that there is virtually no formation of insoluble precipitates of these cations within the resin matrix and on the surface of the beads. However, in a few cases the contents of calcium and/or magnesium were as high as 3.5% and 9% of capacity, respectively. Such irregularities are considered to be indications of defects in carbonatation and purification of regenerant. Contamination of the resin by calcium and magnesium was easily reduced by an acid wash.

Combination of calcium ions in the feed and carbonate ions on the resin can produce turbid liquors. This was observed in tests of the carbonate form of the resin. Normally the concentration of calcium and carbonate ions in liquor after carbonatation corresponds to the solubility product of calcium carbonate. When more carbonate ions are released from the resin the solubility product of $[Ca^{2+}] \times [CO_3^{2-}]$ is exceeded and some calcium carbonate will precipitate. A similar situation occurs with calcium phosphate and the phosphate form of the resin in the case of phosphatation.

Iron and copper are rather dangerous contaminants of the resin. They can form dark-colored complexes both with colorants and with active groups of the resin. As participants in redox reactions they can accelerate degradation and destruction of the resin ion exchange capacity.

Experiments confirmed that, in the presence of iron and copper, the decolorizing performance of the resin deteriorated significantly. This is apparent from the comparison of decolorizing efficiencies of fresh resin in chloride form, and the same resin contaminated by chlorides of iron and copper in acidic solution, followed by reconditioning with alkaline regenerant. The relevant data are summarized in table 2. In all cases the contamination of the resin by these cations resulted in significant deterioration of the decolorizing performance.

It is interesting to note that the mechanism of resin contamination is different for iron and copper, respectively. In the case of iron, the resin acts as a cation exchanger releasing an equivalent amount of hydrogen ions. This means that iron enters the resin either in the form of free ions or cationic complexes.

Uptake of copper is accompanied by reduction of the acidity of the contaminating solution indicating formation of some anionic complexes.

Fresh resin has a limited ability to remove iron from the acidic solution. This ability markedly increases in the presence of colorants on the resin. Copper does not seem to react with fresh resin but again in the presence of colorants the resin is able to accumulate considerable quantities of cupric ions.

TABLE 2--Exchange of cations in contamination of the resin. Effect of cations on decolorizing performance.

Contaminating cation	Cycle No.	Amount of ions adsorbed by the resin, eqv/kg dry resin		Efficiency of decolorization, %	
		H^+	Contaminant	pH 4	pH 9
-	1	0.09 ± 0.09	-	76.5	77.5
	2	-0.16 ± 0.1	-	74.1	74.8
	3	0.08 ± 0.01	-	70.2	65.8
Fe^{3+}	1	-0.18 ± 0.07	0.18 ± 0.03	68.2	63.3
	2	-1.43 ± 0.15	1.56 ± 0.05	67.1	61.1
	3	-1.92 ± 0.08	2.16 ± 0.03	-	-
Cu^{2+}	1	0.00 ± 0.08	-0.07 ± 0.10	74.7	72.2
	2	0.56 ± 0.09	0.41 ± 0.12	63.9	57.5
	3	0.32 ± 0.12	0.36 ± 0.13	-	-

CONCLUSIONS

Resin decolorization of refinery liquors is basically an ion exchange process in which the ionized molecules of colorants replace counter ions initially attached to the resin. Strong base resin and favorable pH of the feed are essential for good decolorizing performance.

Inorganic and low molecular weight organic anions are natural competitors of colorants. In the course of decolorization, these fast-moving anions are able to reach active sites of the resin first. At least part of the exchange of colorants proceeds therefore with these counter ions.

The ionic form of the resin is very important to decolorizing performance. Carbonate and chloride forms are efficient decolorizers whereas resin in silicate and phosphate form is almost inactive. Strong acidic anions on the resin do not significantly influence the pH of liquor, whereas weak anions tend to increase the pH of the product above that of the feed. These changes in pH may influence decolorization and behavior of liquor in further processing.

A high concentration and/or unfavorable composition of liquor ash is a more probable cause of inferior decolorizing of some liquors than peculiarities in composition of colorants. It seems that in such a case a correction could be achieved by selection of a suitable ionic form of the resin and by manipulation of the liquor ash.

Calcium and magnesium can cause reversible deterioration of decolorizing performance. These ions are also percursors of turbidity in decolorized

liquors. Iron and copper are very dangerous contaminants which can cause irreversible damage to the resin.

ACKNOWLEDGEMENT

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DISCUSSION

K. H. Schoenrock (Amalgamated Sugar): With regard to the amount of colorants eluted vs. pH of the regenerant, figure 3 indicates that with decreasing pH

you had less efficient elution. In view of the fact that a strong base exchanger can be regenerated efficiently almost stoichiometrically with a strong acid (HCl) to the chloride form, wouldn't that indicate that the colorants are not really absorbed on the functional sites, but are held by the exchanger by some other form, and that the base that you're using to strip the colorants is merely dissolving them; or, would that indicate that the colorants, being pH sensitive indicators, are changing as they are being eluted so that they are not accounted for. So, to ask my question, did you measure a color balance across the resin? In your last figure, the pH 4 operation indicated actually a slightly better performance in removing the colorant from the liquor, unlike the data in figure 3.

M. Vender: I am glad you mentioned this question because it is extremely interesting and most illustrative. What we found out is that the first step in decolorization is ion exchange and the colorants really are attached to the ion exchange sites. This attachment is temporary and colorants may be released and replaced by anions from liquor and regenerant. Thus there is always a certain proportion of color molecules free to migrate through the resin matrix. We all know that among the colored species are some highly reactive substances, such as for instance tannins, which can easily combine with other molecules to form high molecular weight reaction products. In liquor the molecular concentration of color bodies is low and so is the probability of their collisions and unification. In resin - and I am talking about resins with color accumulation of the order of millions of ICUMSA units - the probability of reactions is much higher. We believe that because of this reactivity colorants are able to create a secondary polymeric structure within the resin phase. This structure is not or only partly attached to the resin so that it is not proper to talk about ionic bonds or adsorption - it seems to be almost exclusively mechanical unification, something like a beast in a cage. Experiment evidence supports our hypothesis. You can extensively regenerate the resin with neutral sodium chloride solution until the effluent is colorless but the resin is still loaded with color. The total ion exchange capacity can be converted to the chloride form but the resin is still like black caviar, has higher density than the fresh resin and its ability to swell is markedly reduced. These are the symptoms of a blockage of resin pores. In this case, a high pH is needed. We need to split the secondary polymer and then, after hydrolysis, the lower molecular weight polymer can get out. The first step is ion exchange, but once you release colorant from this attachment to active sites, it need not necessarily migrate out because it could be connected in some polymeric chain; it could be actually fixed in the empty space of the resin.

K. H. Schoenrock: Are you infering that an acidic regenerant is not a good regenerant for strong base anex? It is, in my experience.

M. Vender: Not in our case, not even 2N hydrochloric acid, which is a pretty strong acid. If you consider regeneration as a conversion of the resin to the chloride form I agree with you that hydrochloric acid is probably the strongest regenerant. But the purpose of our regeneration is also to extract colorants accumulated on the resin. From this point of view the use of acid as the sole regenerant is disastrous. We use, of course, periodical acid washes to dissolve some contaminants such as carbonates, silica and organic complexes of iron.

Another factor must be considered in connection with acidic regeneration. In our experience the decolorizing performance of the resin is best at pH's between 8 and 9. pH of the resin after the acid wash is about 4 so that you need some caustic to bring it to the optimum.

K. H. Schoenrock: So the caustic is stripping the impurities from the functional sites?

M. Vender: Not exactly stripping and not from the functional sites. The effect of caustic is probably twofold. Firstly the high pH supports the hydrolytical fragmentation of the immobilized polymers so that they become mobile again. Secondly it has effect on the molecular shape of colorants. Some substances are like a cluster of snakes, all bent and twisted. At high pH's the molecules straighten out and can more easily diffuse.

R. Moroz (Sucrest): Does that explain why using a strong oxidizing agent such as hypochlorite tends to loosen up or break down the polymer, and open the pores? Have you tried a strong oxidizing agent?

M. Vender: Yes. We have been using hypochlorite. But not in the beginning or middle of the service life of the resin, but at the end because we are afraid that using hypochlorite would not only help to destroy the colorant but would also oxidize and destroy the activity of the resin.

R. Moroz: It destroys the divinylbenzene which begins to break down and makes a soft resin, but it is better than caustic.

M. Vender: I wouldn't say so because we got quite a reasonable result with caustic. We never use caustic as such, we always use alkaline brine. The pH is adjusted to 12.5.

R. Moroz: In using sodium chloride, after you regenerate the resin with sodium chloride, have you ever tried a little hypochlorite in spent sodium chloride to bring it back to reuse it?

M. Vender: We are now trying to do something of this kind, because of the waste water problem.

R. Moroz: In figure 4, you showed the various ionic forms of the resin and their decolorization efficiency. I notice you left one anion out - hydroxyl ion. Have you tried hydroxyl ion from the theoretical point of view?

M. Vender: No. I'll tell you why. In order to convert the resins to hydroxide form you have to treat it with pretty strong caustic and we are working in the range of temperature 85-90°C. To contact the resin with this hot caustic solution would mean that the resin will soon decompose.

R. Moroz: You can cool the resin for the caustic treatment.

M. Vender: All of our cycles are at high temperature.

R. Moroz: I notice you use orthophosphate, have you tried the pyrophosphate or metaphosphate form of the resin?

M. Vender: No.

R. Moroz: The iron and copper contaminants will convert the phosphate to the pyrophosphate form, so, what would happen if you add pyrophosphate? Another question - where did the copper come from?

M. Vender: We were using Monel metal as construction material. This material was not very good for this purpose because during the acid washes the acid dissolved out a bit of copper and brought the copper on to the resin. On one hand the acid was beneficial, but on the other it brought the copper into the system.

M. Matic (S.M.R.I.) Initially you said that this is an ion exchange reaction, but it would appear from the discussion, so it seems to me, that for practical purposes the capacity of the column is made of two things: the ion exchange capacity, and the secondary reaction, which is adsorption. The 1000 bed volumes which you were mentioning, was this just ion exchange capacity or the total capacity of the resin in terms of color elution?

M. Vender: Ion exchange capacity.

M. Matic: So the elution capacity of the resin is considerably greater than that.

M. Vender: Yes. You are right at this point. If we have the resin loaded with colorants which are not attached to the ion exchange sites this additional load would increase the capacity. But this can happen only in the case of cycling through repeated loadings which of course is just what happens in the practical use.

M. Matic: The thousand bed volumes depends upon the raw, and the feed liquor. For practical purposes, obviously you never ran the resin until you completely saturated it.

M. Vender: No.

M. Matic: Then in practice you will be regenerating much earlier. What is the practical cycle length?

M. Vender: Somewhere between 40 and 60 bed volumes per cycle with a life of 200 cycles.

N. H. Smith (C&H): The ratio of colorant exchanged is based on color measurements so presumably in the first cycle this would be a combination of both mechanisms. In table 1, in the second cycle, how do you explain that in two out of the four cases you have a higher capacity on the second cycle?

M. Vender: This ratio depends very much on the composition of colorants. You must realize that, unlike bone char, which after regeneration is more or less back to the original form, the resin in successive cycles is never the same. What could happen easily is that in the first cycle, the resin preferentially removes flavonoids with high indicator value. Then comes the 2nd cycle and because the resin is already considerably saturated with this kind of colorant

it doesn't remove as much. This could actually influence the reading of absorbancy, so it is some indication of the amount of colorant but not really reliable. I reckon that the changes in the selectivity of the resin, cycle by cycle, can explain this effect.

N. H. Smith: On the effect of various ions on pH of solution: if you are going to exchange and liberate something like phosphate or carbonate, the liberated ion would then hydrolyze. Would that account for your high pH?

M. Vender: Yes, that's it.

APPLICATION OF DEIONIZATION PURIFICATION TO PRODUCE LIQUID SUGAR AND SPRAY DRIED PRODUCTS

By Stanley E. Bichsel[1]

ABSTRACT

Sugar beet process thick juice in storage or thick juice direct from the process is purified using two-stage deionization. The purified product when concentrated produces a liquid sugar containing small amounts of invert and raffinose. The liquid sugar product is, however, superior to conventional liquid sugar in terms of decreased color, conductivity ash, and floc content. This liquid sugar is suitable for direct sales or inversion to standard invert products for direct sale. The initial liquid sugar product is also suitable as a spray dryer feed to produce a high quality granular spray dried sugar product. Results of commercial deionization operations and pilot plant runs are discussed with respect to economic feasibility.

INTRODUCTION

Deionization purification of cane or beet sugar liquors has been reported by several investigators (1,2,4,5,7,16)[2]. Most recently a comprehensive review of ion exchange in the beet sugar industry was published by Landi and Mantovani (8). The use of deionization purification has expanded within the sugar industry during the past 25 years. It is estimated that there are now over 25 deionization plants on a worldwide basis. Use of deionization as a means of purifying sugar bearing juices and liquors has increased as a direct function of improvement in ion exchange resins with respect to functional characteristics and physical durability.

The basic economic considerations influencing the profitability of deionization are:

1. Ion exchange plant capital cost
2. Ion exchange plant utility in terms of days of operation per year
3. Ion exchange resin replacement costs
4. Ion exchange regenerant costs
5. Ion exchange sugar produced - sugar in molasses cost differential
6. Ion exchange additional energy requirements as a function of sweetening on-sweetening off water dilution

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[2]Numbers in parentheses refer to items under "References" at the end of this paper.

7. Disposal of high BOD nonsugar impurities eliminated, high BOD resin backwash water, and high or low pH, high salinity, anion and cation regenerant solutions

8. Ease of technical and physical interface with an existent beet or cane mill

In the context of this paper, considerations will be given to these basic elements of economics. This presentation will address the conceptual solutions to basic economic problems and the unique set of circumstances leading to the production of granular spray dried sugar from deionized thick juice.

DEIONIZATION PROCESS

Impurity Removal

Deionization purification is a unique means for purification of sugar liquors because essentially all the impurities, with the exception of nonsugar carbohydrate impurities, are ionized. As ionized molecules in solution, the nonsugar impurities are eliminated as anionic or cationic species. From this general consideration it is evident that the specificity of deionization purification allows complete deashing and decolorization purification to produce a liquid sugar with traces of carbohydrate impurities other than sucrose. In contrast, other less specific methods of purification, based on precipitation of insoluble calcium sucrose salts (Steffens process) (6), separation of sucrose on the basis of molecular size (ultra filtration) (3,15) and separation of sugar from nonsugars on the basis of differences in distribution of highly ionized impurities and weakly ionized molecules, external and internal with respect to the ion exchange resin matrix (ion exclusion) (12,13) are technically inadequate

TABLE 1--Typical ion exchange purification results

	Juice into i.e.	Juice out of i.e.	% Elim.
Refractometric dry substance	27.87	15.50	—
Apparent purity	85.2	95.8	--
Nonsugars x 100/sugars	17.37	4.38	74.8
% (based on sugar)			
(A) Invert	1.42	1.78	--
(B) K	2.02	0.03	98.5
(C) Na	0.73	0.31	57.5
(D) Cl	0.71	0.01	98.6
(E) N	0.95	0.08	91.6
(F) Amino acids + pyrrolidone carboxylic acids	3.52	0.20	94.3
(G) Betaine	0.58	0.19	67.2
Color	1557	135	91.3
pH at 20°C	7.9	8.9	--

in terms of producing quality liquid sugar directly. The basic equipment elements of deionization are described in detail by McGinnis (1). Table 1 shows typical purification efficiencies in terms of percent elimination of nonsugar impurities at the American Crystal Ion Exchange Plant at Moorhead, MN.

Manufacturing Operations

Figures 1 through 5 indicate the conceptual development of deionization in the U.S. beet sugar industry. Figure 1 indicates a standard beet sugar manufacturing factory with allocation of sugar entering as loss 5.0%, sugar bagged for sale 78.0%, and sugar lost in molasses 17.0%. Plain dried pulp produced is expressed as a percent on beets sliced. The values presented are typical industry values and not specific to any one factory in the U.S.

Figure 2 indicates the impact, in increased percent sugar extracted, that the installation of a deionization plant has on sugar production. The utility of such an installation is minimized, however, if the plant only slices beets from 120 to 135 days/year. In addition, this plant incurs a severe liability due to high BOD nonsugar waste discharge, and high and low pH, high salinity, anion-cation waste regenerant discharge. This type of deionization plant was built during the late 40's and early 50's in the United States. At the present time, state and federal laws, developed in recent years, controlling discharge and/or ponding of organic or inorganic pollutants make construction of this type of open waste discharge plant impossible. Treatment costs are prohibitive.

Figure 3 represents, in a conceptual manner, the extension of deionization plant yearly operating time to approximately year-round by utilization of the thick juice storage concept (10). This concept allows expansion of an existing beet sugar factory without capital expenditure to increase the sugar end. This approach to expansion at minimum cost also enhances deionization economics by virtue of increased run time per year. Waste disposal still poses a serious problem in this concept.

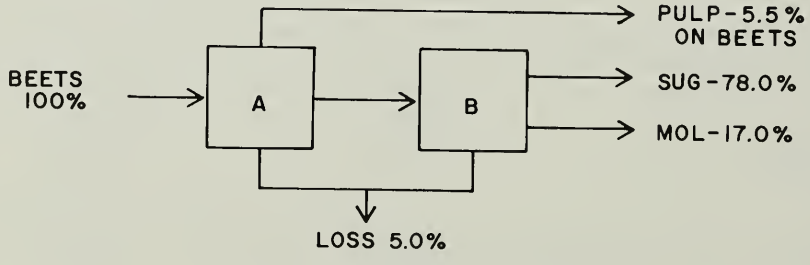

FIGURE 1--Standard factory
A Standard beet end.
B Standard sugar end.

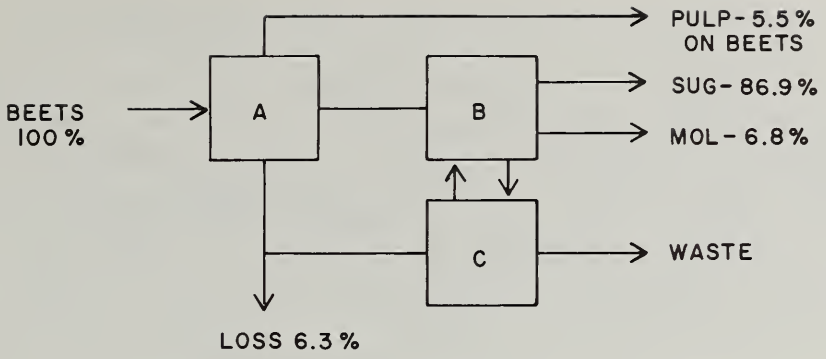

FIGURE 2--Ion exchange factory

C Deionization plant eliminating 60% of the nonsugars entering the sugar end 120 to 135 operating days per year.

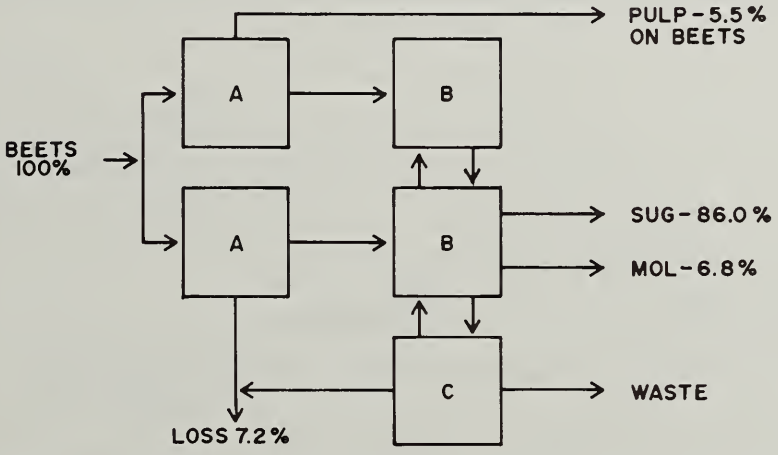

FIGURE 3--Ion exchange factory plus thick juice storage

D Thick juice storage tanks.

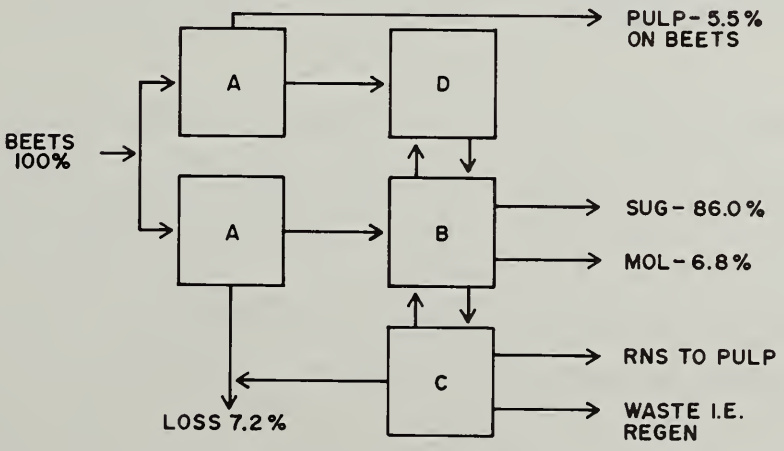

FIGURE 4--Ion exchange factory plus thick juice storage plus reconstituted nonsugar evaporation.

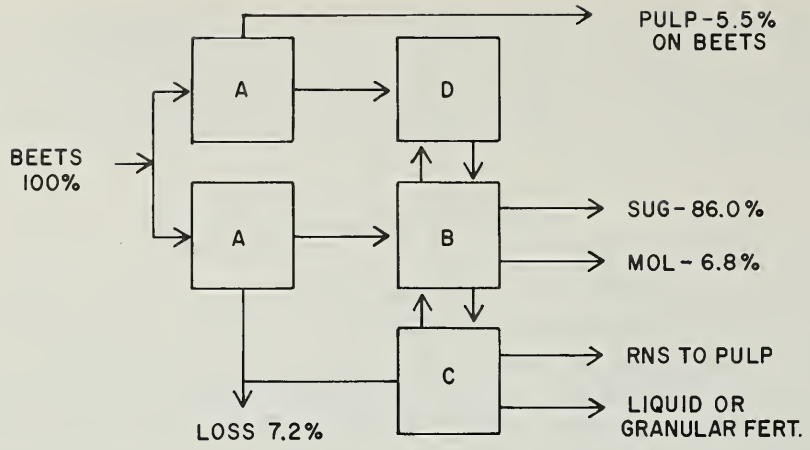

FIGURE 5--Ion exchange factory plus thick juice storage plus reconstituted nonsugar evaporation plus fertilizer production.

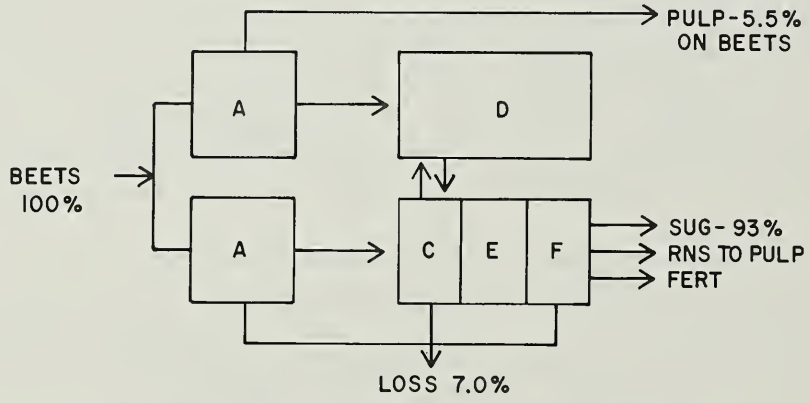

FIGURE 6--Ion exchange spray dry factory
E Ion exchange polishing purification.
F Sucrose spray dryer.

Figure 4 duplicates figure 3 with a significant exception involving a partial solution to waste disposal (2,9). In this case R.N.S. (reconstituted nonsugars) are concentrated and diverted to pressed beet pulp prior to drying. Nonsugars eliminated during deionization purification are, by and large, nitrogenous organics in nature, amino acids and betaine. The protein content of the dried beet pulp was thus increased approximately 20%. This approach to waste stream conversion to a profitable by-product credit solved a serious technical waste disposal problem. Failure to solve this problem would have stopped consideration of further deionization plants on the basis of prohibitive waste treatment costs.

Figure 5 indicates the present state of the art of deionization purification in the U.S. beet sugar industry. This concept differs only from figure 4 in that the regenerant waste streams are concentrated to produce a homogeneous, granular, fertilizer with a formula of 17-0-5-19. The fertilizer is produced by a spray dry, fluid bed, granulation process on a continuous basis. The recently commissioned American Crystal Moorehead, Minnesota, Deionization Plant produced up to

48 short tons of granular fertilizer per day and approximately 35 short tons of R.N.S. on a dry solids basis per operating day. This is the first plant in the foreign or domestic beet sugar industry to produce a granular fertilizer by-product. The economic impact of applying fertilizer by-product credits back against initial regenerant costs is highly significant. The production of by-product R.N.S. and a liquid fertilizer are covered under U.S. Patent No. 3,700,460 (2). Techniques and specialized equipment required to produce a granular fertilizer from waste regenerant concentrates are classified as proprietary at this time. Table 2 indicates ion exchange incremental sugar production cost-credit analysis, with emphasis on by-product credit impact on the production cost per hundred pounds of deionization sugar.

TABLE 2--Ion exchange incremental sugar production cost-credit analysis

Assumptions	
Sugar at $13.50 (net)	
Molasses 79.5 Bx at $60.00/ton	
Pulp at $60.00/ton	
Plant operating at 80% design capacity	
Plant operating 350 days/year	
Product Credits	
Sugar cwt: 412,160 at $13.50 net	$5,564,160
R.N.S.(Reconstituted nonsugars) at 79.5 Bx at $60.00/ton 15,391	$ 923,460
Fertilizer 17-0-5-19. 16,000 Tons at $91.11/ton	$1,466,834
Gross credit/year	$7,954,454
Operating Costs	
Chemicals (regenerants, etc.)	$1,508,634
Resin replacement & make-up	$ 225,207
Utilities (energy & water)	$ 458,440
Labor (R & M)	$ 569,297
Molasses (not produced & shipped & freight)	$2,141,130
Gross cost/year	$4,902,708
" " + depreciation	$5,286,041
Before tax credit/year	$2,668,413
Cost of production/cwt (no. B.P. credit)	$12.83/cwt
R.N.S. B.P. credit	$ 2.24/cwt
Fertilizer B.P. credit	$ 3.56/cwt
Total B.P. credit	$ 5.80/cwt
Cost of production/cwt (with B.P. credit)	$ 7.03/cwt

TABLE 3--Typical liquid sugar quality as produced by two-stage deionization purification of thick juice

	Thick juice to primary ion exchange	Thick juice to secondary ion exchange	Type - 0 liquid sugar
Apparent purity	91.3	98.4	98.6
% Invert on dry substance	0.62	1.21	1.32
% Raffinose on dry substance	0.40	0.42	0.39
Total N	0.70	0.07	--
Na	0.42	0.14	--
K	1.02	0.001	--
Cl	0.42	0.13	--
Color	728	290	18
pH	9.0	8.4	7.2
Cond. ash	--	--	0.005
Floc, ppm (beet sugar test)	1100	0.5	--

Figure 6 indicates the concept involved in the direct year-round production of liquid sugar and/or spray dried granular sugar from two stage (primary and secondary) polishing deionization of process or stored thick juice. Techniques involved in producing liquid sugar directly have been reported in the literature (15). In this case, thick juice purified with the conventional strong cation-weak anion is further purified with a strata-bed strong anion-strong cation to produce a high quality liquid sugar. Potassium hydroxide is utilized to regenerate the strong anion waste. Regenerant solutions made up of ammonium sulfate and potassium sulfate are concentrated and processed to a granular fertilizer as is done at present at the new Moorhead Deionization Purification Plant. Liquid sugar may be sold directly or spray dried to a granular product. Table 3 indicates the quality factors in purification of thick juice to Type-0 liquid sugar grade using primary and secondary deionization purification. Table 4 compares spray dried sugar, as produced from deionized thick juice, with regular fine granulated sugar. Specific techniques and the specialized equipment involved in the production of spray dried sugar are proprietary at this time with several patents applied for. Direct production of liquid sugar, by deionization of thick juice followed by spray drying to produce a high quality granular product, eliminates the sugar end as it now exists. Vacuum pan boiling, centrifugal separation of mother liquor and crystal, and long-retention-time crystallizers are replaced by a straight-through no-recycle deionization purification of thick juice, followed by spray drying.

CONCLUSIONS

An overall assessment of spray dry economics, taking into consideration capital requirements, energy requirements, potential labor reduction through automation of a continuous sugar production process, and positive economic impact of maximum sugar extraction and fertilizer and R.N.S. credit, indicate that the

TABLE 4--Comparison of fine granulated sugar with spray dried sugar

	Regular fine granulated	New product
Purity	99.8% (min)	98.0 (min)
Color (RBU)	20 to 35 (max)	5 to 15 (max)
Ash (conductivity)	0.015 (max)	0.005 (max)
Floc (saponin)	5 ppm (max)	none
Sulfur dioxide	6 ppm (max)	3 ppm (max)
Oligosaccharides (primarily raffinose)	0.1% (max)	0.5% (max)
Monosaccharides (glucose & fructose)	0.05% (max)	1.5% (max)
Sediment, ppm	3 ppm (max)	3 ppm (max)
% Moisture	0.050	0.050
Bulk density (lb/cu ft)	53 - 55	48 - 50
Screen analysis (accumulative % retained)		
+ 28 Tyler	1% (max)	5% (max)
+100 Tyler	99% (max)	50%
+150 Tyler	100%	82%
+200 Tyler	100%	98%
Pass - 200 Tyler	trace	2%
Physical description	white, well-defined crystals	white, amorphous particle structure
Odor	faint storage odor	none

spray dried sugar process has high potential for future use in the sugar industry. The process is the culmination of technology developed over the past 30 years in the areas of thick juice storage, improvements in resin, deionization hardware, conversion of deionization waste streams from liabilities to by-product credits and development of advanced spray dry technology. All these advances were necessary to bring the industry to the present state of technological awareness.

Continued effort, time, and capital will be necessary to implement the concepts described in this paper on an industry-wide scale. Economic necessity is the mother of technological change. In the competitive world of sweetener production, continued existence without recourse to technological change will be, at best, perilous.

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DISCUSSION

K. R. Hanson (Amstar): To avoid a stampede to the microphone, would you explain how the beet industry gets 105% sugar out for 100% in? It involves the difference between beets sliced and beets purchased, and covers the sugar in crowns.

S. E. Bichsel: We don't calculate extraction in that manner. Extraction is based on sugar in beets introduced compared with sugar produced.

K. H. Schoenrock (Amalgamated Sugar): Concerning the bulk storage of spray dried sugar: have there been pilot studies along this line, and, if so, what were the results of those studies?

S. E. Bichsel: We did conduct tests over a duration of six months in a temperature-humidity controlled room. We tried to simulate the pressure found at the bottom of a silo on both mill-run granular sugar as a standard, and spray dried sugar, as a comparison. We cycled the atmosphere surrounding the sugar with high temperatures and high humidity and then cold temperatures. After a six-month test, the regular granulated sugar set up literally like a brick; the spray dried sugar had only a few small lumps, maybe the size of your fingertip, that were friable - you could crumble them with your fingers very easily - but, essentially, it did not set up at all. It remained free flowing.

K. H. Schoenrock: What would happen if the spray dried sugar were to contain various concentrations of invert components such as fructose which is highly hygroscopic?

S. E. Bichsel: This is something that we have just investigated somewhat cursorily, and this could cause problems. We don't know enough about it yet. We've got to run more tests on the high invert-containing spray dried sugar.

J. D. Lewis (Westcane): In using the strong cationic resin followed by the weak anionic do you get consistent measurement performance of ash removal for total cubic foot of resin? If so, what is the ash measurement that you get?

S. E. Bichsel: I can't give you that figure without some calculations. We watch very closely the tons of sulfuric acid regenerant and the tons of anhydrous ammonia regenerant used per ton of nonsugar impurities eliminated. This, in conjunction with gallon through-put, cycle lengths, and number of regenerations per day gives us an idea of whether our efficiency is dropping off. I could calculate this if I knew the average molecular weight of our impurity, which I could assume, but I don't have the true value available.

J. D. Lewis: The figures that you showed were an input ash 0.15%, an output ash of 0.006%. Could you give any indication, using those figures, of the duration of a service cycle of a column? I know, of course, that it depends on the size of the column.

S. E. Bichsel: We get about four and a half to five hours on an anion column; we run them chock-a-block or merry-go-round, in the secondary position. The fresh resin going on is the last resin to see the juice. Then, as it's rotated into the primary position where it is fully exhausted, a column that was in the primary position goes into regeneration. You could say that we have an anion in service somewhere between eight and nine hours, including secondary service first, and then primary service. Now, on the cation, when we are running close to design capacity, we have about two-hour service cycles.

C. H. Novotny (Industrial Filters): One of the big differences between the beet operation and a cane refinery is the removal of tons and tons of nonsugars that are not necessarily ash from beet juice, while the bulk of the cane refiners ion exchange load is ash.

S. E. Bichsel: I think of that in terms of the applicability of the spray dry concept to the cane people, particularly to the cane organizations that take raw cane sugar, clean it up with carbon and deashing and decolorization, and then sell liquid sugar. It seems to me that going on to spray drying would be a natural thing for them. This approach would enable the producer of liquid sugar directly from raw sugar to expand the product line to granular with minimum capital outlay.

K. H. Schoenrock: The concept of spray drying and total ion exchange as presented undoubtedly was considered for your Moorhead expansion, and you still decided to go the conventional way. What was the motivating force? Why didn't you go to spray drying?

S. E. Bichsel: For two reasons: we do have fairly serviceable and relatively new conventional sugar end equipment at Moorhead. For example, three years ago they put in a new white centrifugal station. We feel we just can't tear this equipment out and sell it on the market at fifty cents on the dollar. It has to be amortized. The other reason is, we do feel that spray drying is still somewhat experimental and we'd like to do it on a day-in, day-out pilot basis before we would consider recommending it to anyone or using it ourselves.

K. H. Schoenrock: Would you say then that the second reason is the primary reason, because with many plants there would certainly be a place to put this new centrifugal station? I just wondered how much each of these factors weighed in the final decision. Was it primarily the newness of the process?

S. E. Bichsel: Another thing is that our technical people have had their hands full working out problems with the primary deionization plant. Even though these plants have been built before, we have had to train people, and in addition, we have had some problems in working out production of granular fertilizer. To add on to that a secondary purification in spray drying would be a rather difficult thing for people to assimilate, so the decision was a combination of two things. I think it is good to learn to walk before you learn to run.

C. C. Chou (Amstar): The invert content of the spray dried sugar seems to be rather high. What would be industrial user's attitude toward this type of sugar with invert content of 0.5 to 1%?

S. E. Bichsel: Customer acceptance of spray dried sucrose containing invert levels ranging from 0.5 to 1.0% are dependent on the product produced. As an example, liquid sugar (Type 0) sold to or produced by bottlers may be used to produce soft drinks which are acidic. Natural inversion occurs as a function of shelf life time. Invert percentages mentioned would have little effect on soft drink quality.

C. C. Chou: What would be the keeping quality of spray dried sugar, particularly with regard to hygroscopicity?

S. E. Bichsel: As I mentioned before, the spray dried sugar does not cake very much at all. Apparently the surface area of spray dried sugar has a greater capacity to assimilate moisture resulting from cooling warm, high humidity air than the solid crystalline sugar. Crystalline sugar has relatively flat crystal surfaces which can act as condenser surfaces and surfaces for sticking one crystal to another when the crystal surfaces have traces of moisture present. Spray dry sugar, on the other hand, has a heterogeneous surface made up of many micro crystals. The spray dried particle is porous throughout and presents a somewhat larger internal condenser surface area than conventional crystalline sucrose.

C. C. Chou: Do you foresee consumer (grocery product) acceptance of spray dried sugar with different product characteristics and/or form as compared to conventional granulated sugar?

S. E. Bichsel: Spray dried sugar is finer than conventional sugar and does not have the sparkle associated with the crystallized sugar. It presents a flat white powder appearance similar to powdered sugar. Although spray dried sugar does not have the sparkle that crystalline sugar does, it has certain advantages to the housewife in the rate of dissolution. Spray dried sugar dissolves instantaneously in iced drinks, cake mixes, candy (such as fudge), and cake icing - problems with undissolved grain are nonexistent. The bulk density of spray dried sugar is 50 lb/cu ft which approximates the 53 to 55 lb. range most recipes are designed around. The housewife might have to adjust spray dried sugar volume measurement by 5 to 10% to accommodate certain recipes. All things considered, I believe spray dried sugar would be accepted by the grocery trade after a certain market induction period.

C. C. Chou: What industrial market segment would use this type of spray dried product.

S. E. Bichsel: Spray dried sugar could be utilized by industrial customers in applications where grinding is required, and where high rate of dissolution is important. In the case of spray dried sugar produced from liquid sugar derived from deionization purified beet thick juice, the spray dried sugar produced is totally devoid of floc-producing nonsugar impurities. Those considerations and others which are of a proprietary nature indicate that spray dried sugar will occupy a significant niche in the industrial market place in the future.

DETERMINATION OF CADMIUM, ZINC, LEAD, AND COPPER IN REFINED SUGAR BY AN ELECTROANALYTICAL TECHNIQUE

By R. Cormier, L. H. Mai, and A. D'Antico[1]

(Presented by R. Cormier)

ABSTRACT

The determination of metal elements in sugar products usually requires tedious preparations that may give rise to a loss in the accuracy of the results.

DPASV (differential pulse anodic stripping voltammetry), coupled with a rotating mercury film electrode, may overcome this problem. Such analysis can be done, down to a sub-ppb, level without any prior ashing, ion exchange or other similar treatments.

INTRODUCTION

In a previous paper (14)[2] it has been shown that the differential pulse anodic stripping voltammetry (DPASV) technique coupled to a hanging mercury drop electrode (HMDE) can be employed for the detection of trace elements in sugar products, at less expense than for many other instrumental devices. As was reported in work on the determination of lead and copper in raw sugar, (14) it has also been found that a mercury film electrode (MFE) in place of the HMDE could be of great interest when considering higher resolution and sensitivity.

The sensitivity of the DPASV method increases when coupled with a rotated MFE rather than an HMDE. The increase in the magnitude of the signals (peaks current) is the result of an enhanced mass transport, and of a uniform layer of mercury of small thickness on the surface of the electrode. Unlike the HMDE, the MFE exhibits almost no diffusion phenomena into the mercury electrode itself.

Nevertheless, although the sensitivity and the resolution are better, the accuracy suffers from the switch to a rotated MFE, because the surface of that electrode becomes rapidly contaminated with adsorbed or electrodeposited materials and requires frequent cleanings.

INSTRUMENTATION

The study was made using a Princeton Applied Research (PAR) polarograph, model 174, to which was coupled, for some tests, a PAR automated electroanalysis controller model 315.

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[2]Numbers in parentheses refer to items under "References" at the end of this paper.

A three electrodes system was used. The working electrode was a Beckman glassy carbon rotating electrode, #39084, (surface area approx. 1.1 cm^2), powered by a Beckman rotating assembly, #188501. Potentials were measured against a saturated calomel electrode, put in contact with the solution by means of a salt bridge tube incorporating a Vycor tip to minimize leakage. The counter or auxiliary electrode was a platinum wire.

The polarograms were recorded on a Brinkman potentiometric recorder, Servogor 2S.

A chemical reducing solution, in conjunction with a filter, was used to remove oxygen traces from the stream of argon used to purge the analyzed solution of dissolved oxygen. The whole electrolytic cell was kept at a constant temperature of 30°C in a water bath. Standard additions of metals were performed with Gilmont microburettes, of 2500 µl and 250 µl capacities.

ANALYTICAL PROCEDURE

The sugar solution for analysis was prepared by dissolving the sugar sample into the selected electrolyte; in the case of refined white sugar, a 10% w/v solution was prepared.

The selected electrolyte was 0.3M distilled triethanolamine (B.P. 150° to 155°C/0.5 mm to 0.6 mm Hg) and 0.1M sodium hydroxide; the pH was adjusted to 8.5 with concentrated nitric acid (Aristar grade, BDH). According to Barendrecht's procedure[2], the deionized water was distilled before use (conductivity: 1.65 µmhos).

For reasons given later, treatment of the cells with alkylchlorosilane reagents[11] should be avoided. All the glass instruments are washed with NaOH, 1M, to prevent cadmium adsorption.

The analyses were carried out as follows: a 20 ml aliquot of the sample solution was pipetted into the electrolytic cell. To this solution was added 200 µl of a 2000 ppm Hg^{++} solution, to allow the in situ plating of the mercury film. The solution was then purged with argon for 10 min; at the end of that period, a potential of +0.6 V was applied for 3 min in order to condition the working electrode.

Next, the deposition was performed for 5 min at a fixed potential of -1.6 V or -0.9 V while the electrode was kept rotating at a speed of 2400 rpm.

Rotation was stopped, and the system left standing for 30 sec, after which an anodic scan was initiated from the deposition potential to a final potential of +0.6 V, at a rate of 10 mV/sec. During the scan, pulses of 25 mV in amplitude are applied to the ramp of voltage every half second. The entire procedure was repeated twice.

After the second scan, the solution was spiked with known amounts of the element or elements of interest by means of a microburette; each addition was followed by a 2 min purging of the solution before the usual sequence of analysis was begun.

The technique used requires four basic steps:

1. Conditioning of the working electrode
2. Deposition of the metals and of the mercury onto the surface of that electrode (reduction process)
3. Equilibration (static step)
4. Anodic stripping of the deposited materials from the surface of the working electrode (oxidation process).

RESULTS AND DISCUSSION

In earlier work in this laboratory[1] an HMDE was used; however, since this electrode suffers from a low ratio of surface to volume, an MFE was substituted. Although this is also a mercury electrode, it has a very small volume. The thickness of the mercury film has been reported to be in the range of few angstroms[4,9].

There are two possibilities when considering the preparation of the mercury film on the surface of the MFE. The electrode can be plated outside the cell[4,5,18], but that has the disadvantage of introducing another step into the analytical procedure. In order that analysis remain as simple as possible, an *in-situ* plating of the mercury, as suggested by many workers[4,9,11,12] was done. Studies were undertaken to optimize the concentration of the mercuric ions into the electrolyte with regard to sensitivity and resolution, and 1×10^{-4} M was found to be a desirable concentration. The MFE was rotated in order to improve mass transport in the neighborhood of the electrode, and also resolution. A speed of 2400 rpm was found to serve those objectives best; higher speed would have been desirable, but splashing occurred at higher speeds.

The supporting electrolyte was chosen to avoid, as far as possible, any sample pre-treatments such as ashing[13] but the solution prepared contains large amounts of sucrose. Sucrose is known to complex metallic ions; consequently, it was imperative to avoid using an acidic electrolyte in order to prevent any chemical inversion of the sucrose. As an alkaline electrolyte makes the determination of iron easier, basic electrolytes known to afford analysis of iron were thus considered. On consideration of tartrate and triethanolamine[16] based electrolytes, the latter was chosen for its better resolution and for the easiness with which it can be purified. But the search for an electrolyte affording the analysis of iron under the conditions used was unsuccessful.

Figure 1 shows the kind of polarogram obtained when depositing at -1.6 V. Table 1 summarizes the characteristics of the stripping potentials. To the list, in table 1 iron and nickel species should be added. Unfortunately, these species were found to show such erratic behavior that only qualitative determinations are possible and, even then, at a ppm level. That behavior can be explained by the fact that those species do not form amalgams upon deposition onto the surface of the MFE, but rather from hydroxides[3]. In such cases, a DME whose surface is continuously renewed would be more appropriate for analysis.

After a study of the working parameters, a sample of refined cane sugar was analyzed for its copper, lead, cadmium and zinc content. The determinations were achieved by the standard addition method.

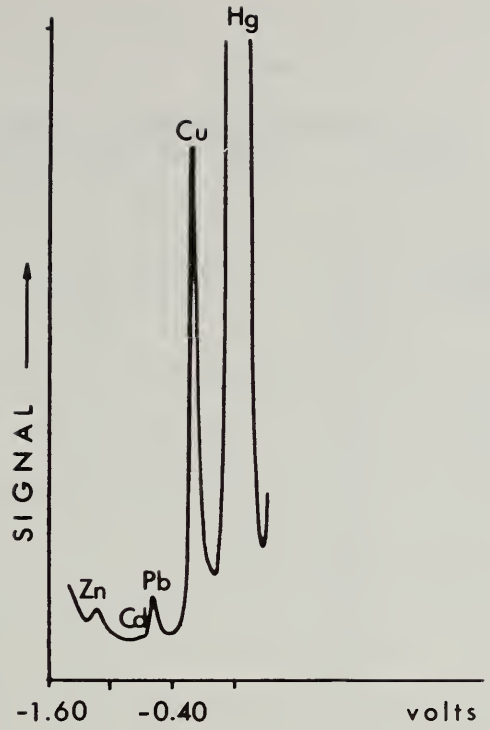

FIGURE 1--Polarogram obtained when depositing at -1.6 V, with supporting electrolyte: 0.3M triethanolamine, 0.1M NaOH, pH 8.5.

TABLE 1--Stripping potentials for various ions

Specie	Stripping potential vs SCE (V)
Zn	-1.14
Cd	-0.74
Pb	-0.60
Cu	-0.22
Hg	+0.21

Figures 2 and 3 show respectively the polarograms obtained for the determinations of lead and copper in both the electrolyte and the sample solution; cadmium was not detectable. Figure 4 demonstrates the analysis of the three species simultaneously. As can be seen in all these polarograms, the baseline is relatively satisfactory.

Table 2 summarizes the results obtained. However, the quantity of copper determined was very doubtful, for reasons which are given later. A colorimetric analysis [15] showed 183 ppb copper, and confirmed the unreliable result by DPASV.

TABLE 2--Analysis for lead, copper, and cadmium

Trace metals		Concentration of trace metals, ppb		
		Blank	10% Refined sugar	Refined sugar
-1.6 V	Pb	9.1 ± 0.8	11.8 ± 1.2	27
	Cu	26.0 ± 3.9	70.8 ± 7.6	448
	Cd	< 0.2	< 0.2	< 1
-0.9 V	Cu	23.5 ± 2.3	41.2 ± 3.9	177
Copper by colorimetric analysis				183 ± 3

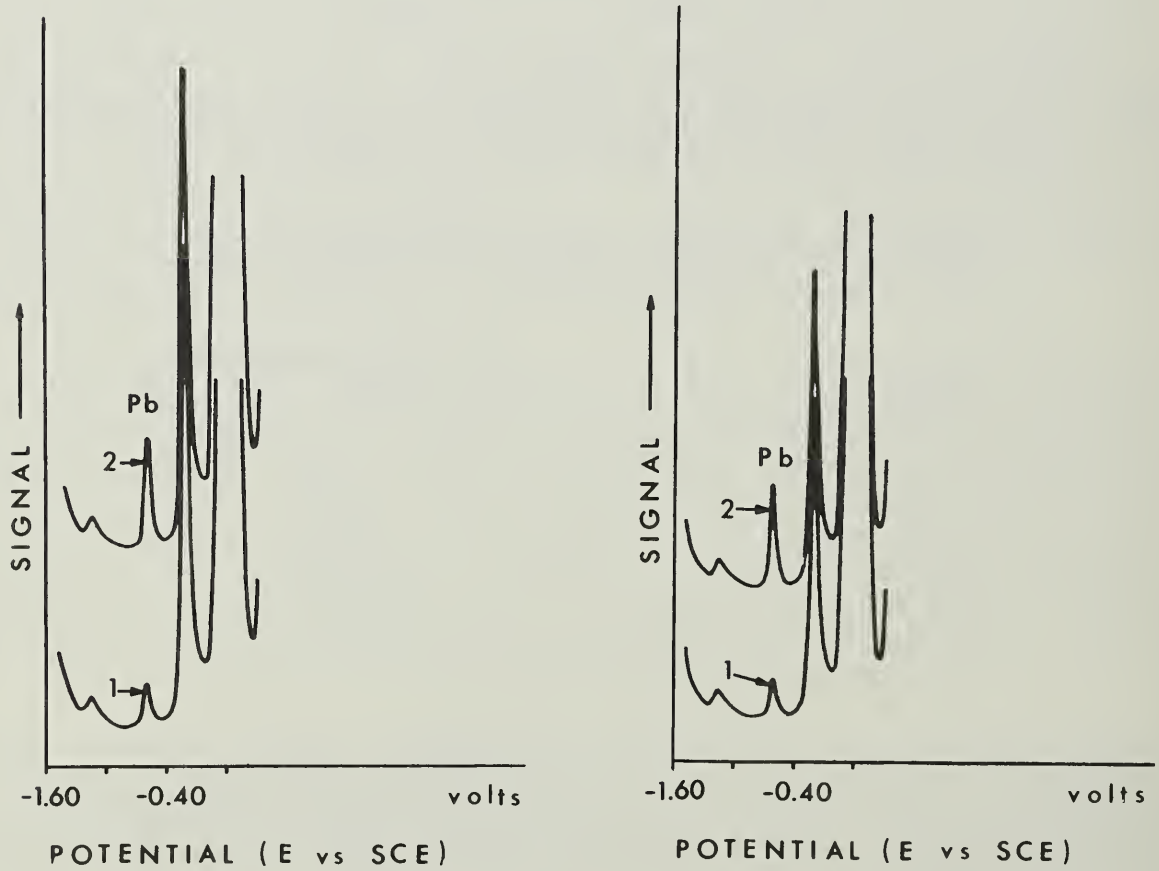

FIGURE 2--Determination of lead.

In electrolyte.
(1) 20 ml 0.3M triethanolamine, 0.1 M NaOH, pH 8.5
(2) as in (1) +8.1 µl 20 ppm Pb^{++}

In sugar.
(1) 20 ml 10% sugar in triethanolamine buffer
(2) as in (1) +10.0 µl 20 ppm Pb^{++}

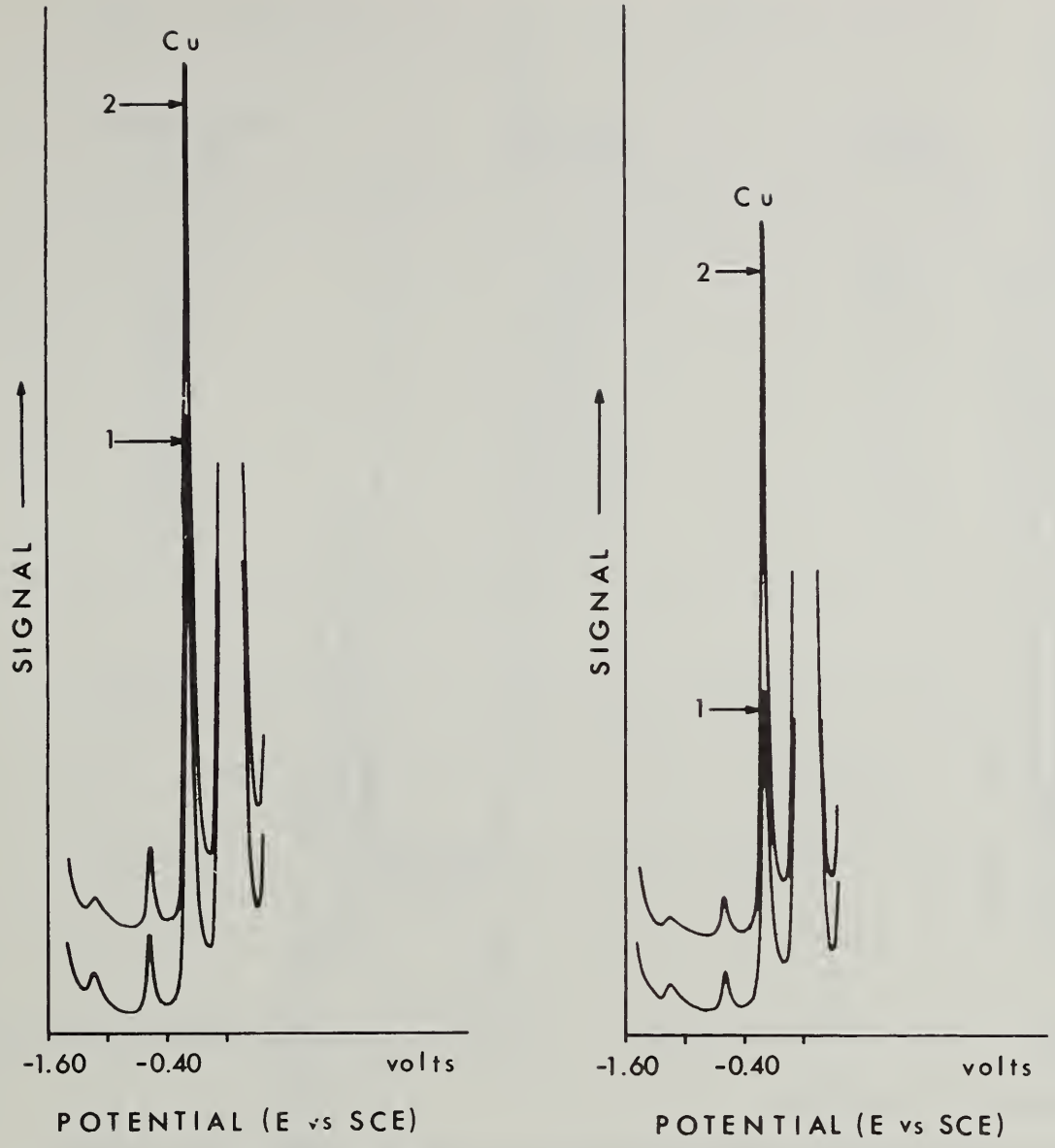

FIGURE 3--Determination of copper.

In electrolyte.	In sugar.
(1) 20 ml 0.3M triethanolamine, 0.1M NaOH, pH 8.5	(1) 20 ml 10% sugar in triethanolamine buffer
(2) as in (1) +1 µl 200 ppm Cu^{++}	(2) as in (1) +7 µl 200 ppm Cu^{++}

It was noticed that, upon variation of the relative concentrations of the elements in the analyzed solution, whereas peaks for lead and cadmium remained stable, the peak heights for zinc and copper varied. Such behavior has been attributed by many workers to the formation of intermetallic complexes between copper and zinc species within the amalgam[4,6,7,10,17]. Following Copeland's suggestion[6], gallium ions were added to the solution in order to prevent interactions between copper and zinc. This was ineffective, probably because of the alkaline pH of the electrolyte.

Before attempting to achieve a better understanding of the problem with copper and zinc, copper analysis was done by changing the potential to avoid the

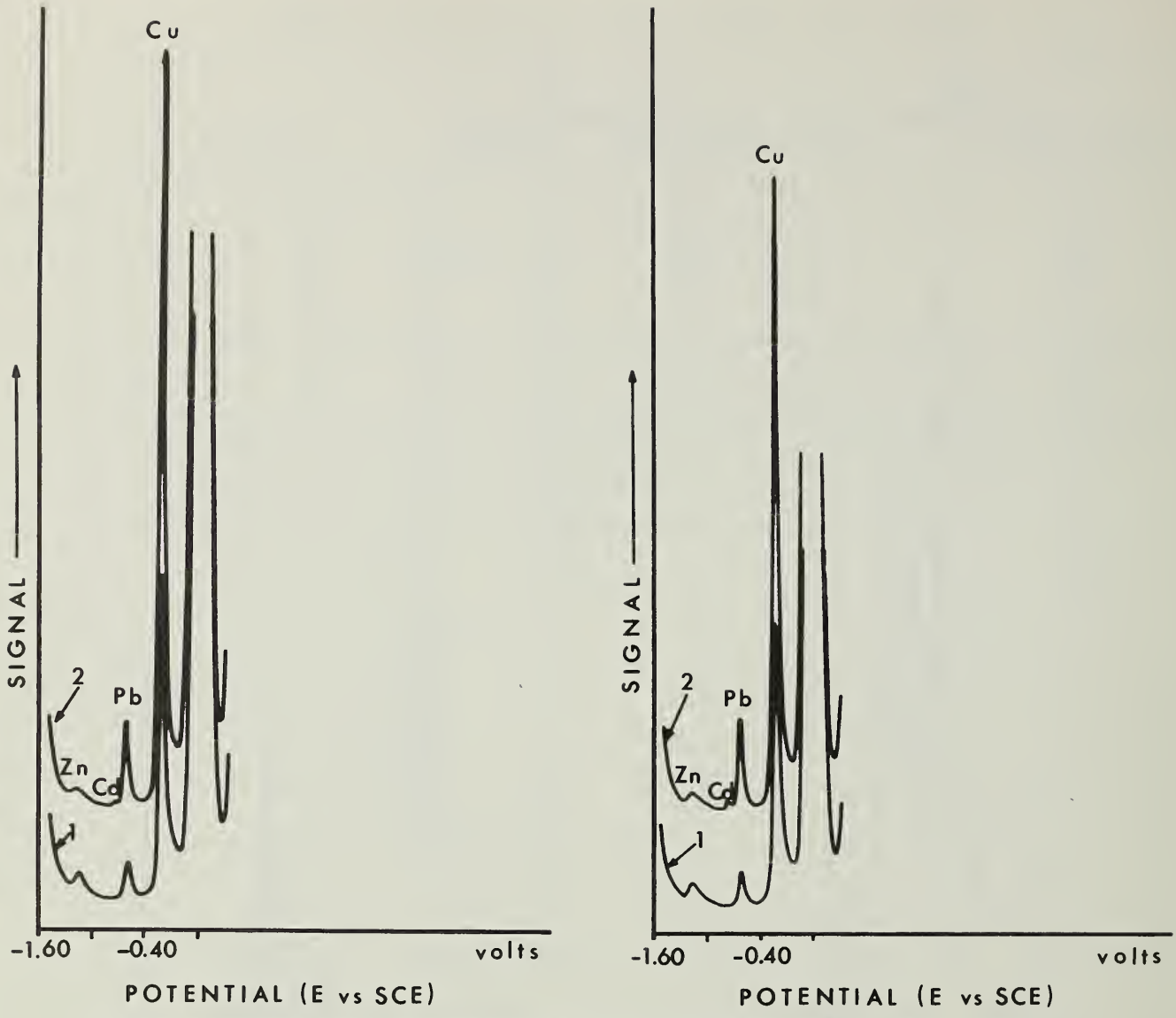

FIGURE 4--Simultaneous determination of copper, lead, and cadmium.

In electrolyte.	In sugar.
(1) 20 ml 0.3M triethanolamine, 0.1M NaOH, pH 8.5	(1) 20 ml 10% sugar in triethanolamine buffer
(2) as in (1) +2.0 µl 200 ppm Cu^{++}	(2) as in (1) + 7.0 µl 200 ppm Cu^{++}
+7.08 µl 20 ppm Pb^{++}	+10.0 µl 20 ppm Pb^{++}
+0.4 µl 20 ppm Cd^{++}	+ 1.0 µl 20 ppm Cd^{++}

deposition of zinc. This was best done by using a potential more positive than the reduction potential of zinc; the deposition was performed at -0.9V. The electrolyte was then found to have a copper content of 23.5 ppb while the 10% sugar solution had a content of 41.2 ppb; therefore, the copper content of the refined sugar was 177 ppb (see figure 5). That number is much more consistent with the result obtained from the colorimetric analysis. However, the determination of zinc with the electrolyte and sugar solutions still remains a problem.

Seeking a simple solution to the problem, the effect of copper on the peak height of the zinc specie was studied. According to the work by Shuman et al, (17) 2000 ppb of zinc were added to the electrolyte which was then titrated with copper (bearing in mind that the electrolyte already contains 23.5 ppb of copper).

TABLE 3--Titration of a 2 ppm Zn^{++} solution with Cu^{++}

Copper added (ppb)	Height Cu (mm)	Height Zn (mm)
0	23.5	100
200	84.5	100
400	100	97
800	116	91
1200	122	47
1600	133.5	41
2000	139.5	28
2400	144.5	15.5
2800	148.0	11.5

TABLE 4--Titration of 2 ppm Cu^{++} solution with Zn^{++}

Zn added (ppb)	Height Cu (mm)
0	85
400	90.5
800	91
1200	83
1600	77
2000	75
2400	68.5
2800	58

The peak heights were monitored for zinc and copper. Figure 6 illustrates the results given in table 3. It is interesting to note that another peak adjacent to zinc, but not completely resolved from it, decreases in height when copper is added to the 2 ppm zinc solution. On the other hand, the height of that peak increases when zinc is added to a 2000 ppb copper solution. In the latter case, the peak height for zinc became difficult to monitor and hence is not included in figure 7 which illustrates the results of that experiment, given in table 4. As can be seen from figures 6 and 7, two curves are obtained, with a "plateau"; Shuman obtained two intercepting straight lines in similar work with a 2 ppm zinc solution.

Before these results were examined, a check was required on the direct proportionality between the peak heights and the concentrations of the copper and of the zinc species. Using the electrolyte as is and assuming a linear relationship of the type, Peak height = k x concentration, the peak height for the zinc specie was plotted vs the amount of zinc added to the electrolyte. Figure 8 illustrates the result. The graph indicates that at a concentration of 2000 ppb of zinc, some kind of saturation problem is occurring.

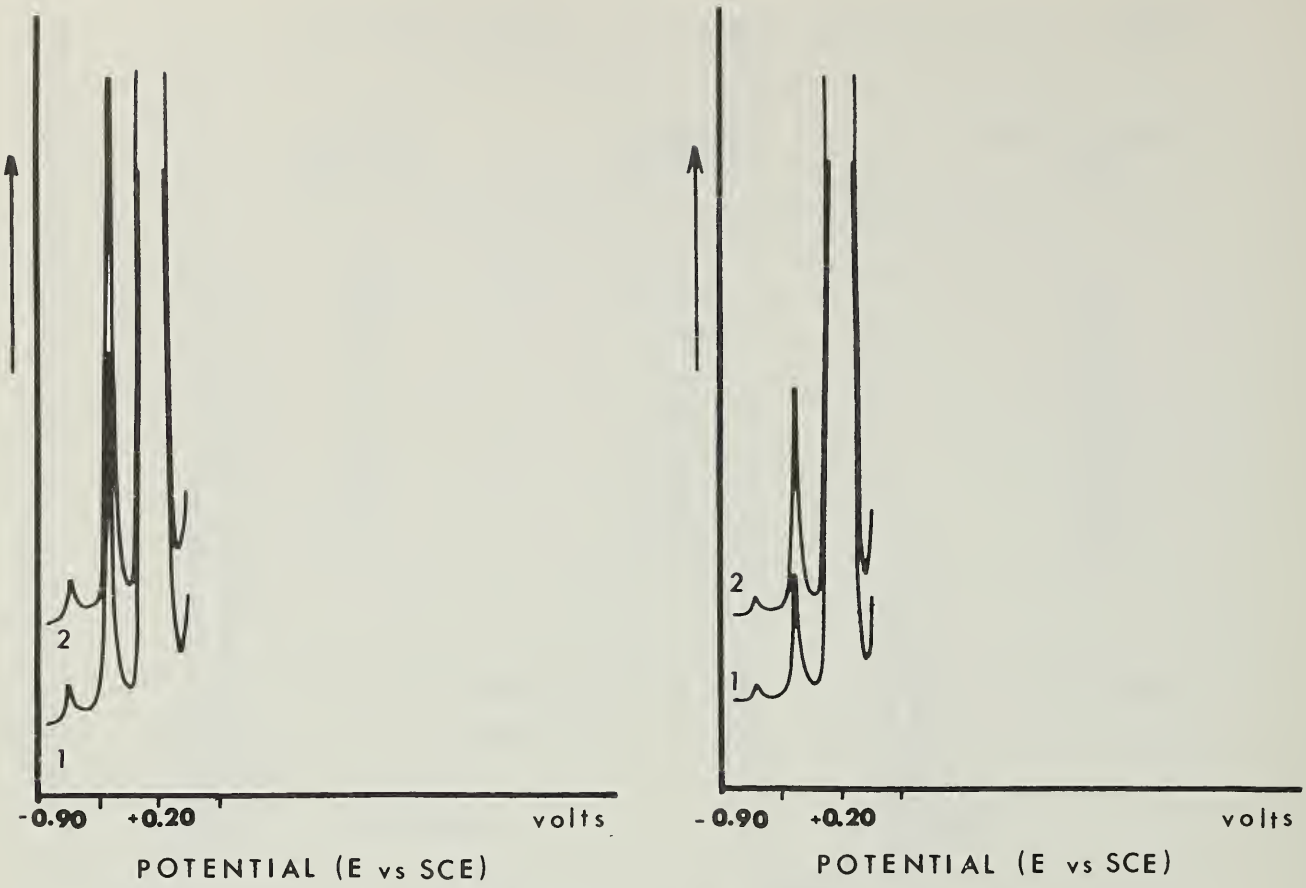

FIGURE 5--Determination of copper without interference from zinc by depositing at -0.90 V.

In electrolyte.
(1) 20 ml 0.3M triethanolamine, 0.1 M NaOH, pH 8.5
(2) as in (1) +10 µl 20 ppm Cu^{++}

In sugar.
(1) 20 ml 10% sugar in triethanolamine buffer
(2) as in (1) +30 µl 20 ppm Cu^{++}

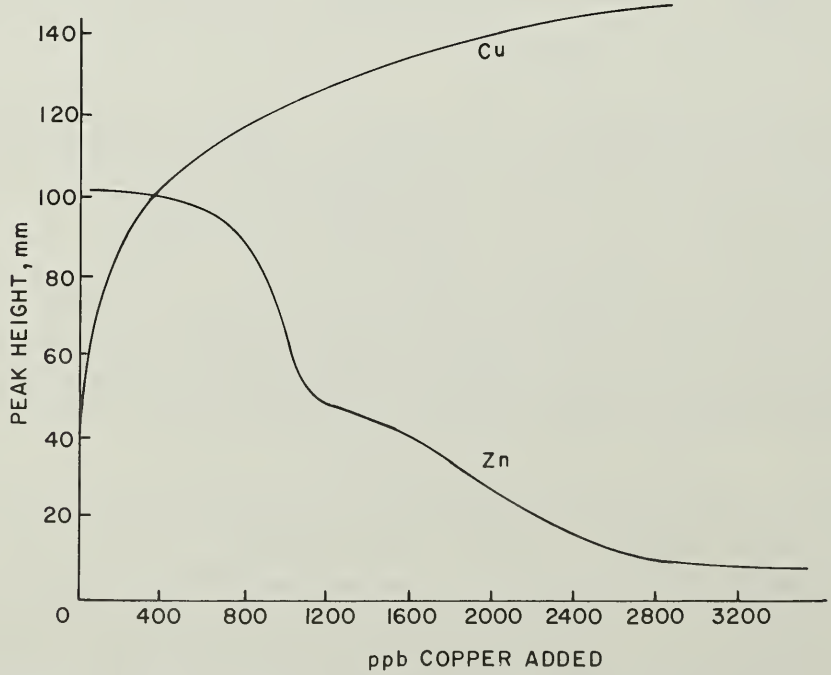

FIGURE 6--Peak height vs Cu added in presence of 2 ppm Zn.

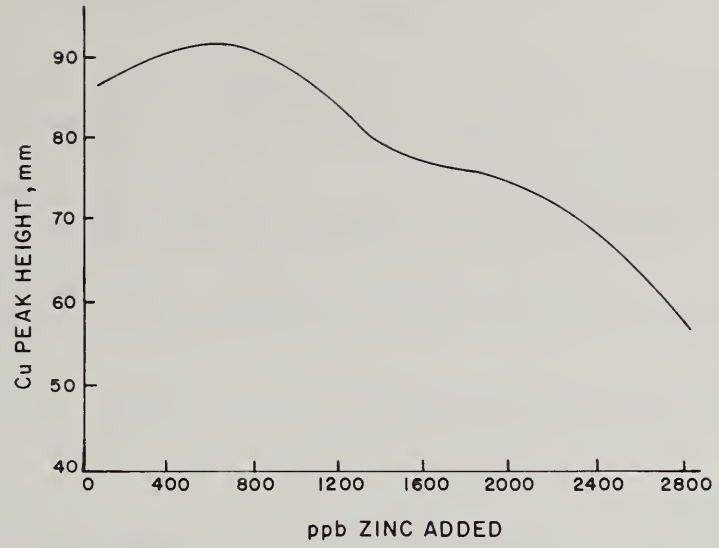

FIGURE 7--Cu peak height vs Zn added in presence of 2 ppm Cu.

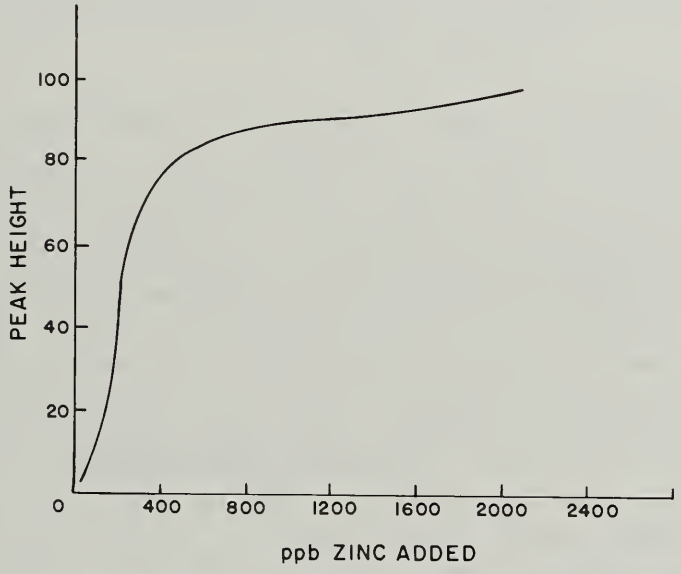

FIGURE 8--Zinc peak height vs zinc added.

Another titration of zinc with copper was then performed but this time only 150 ppb of zinc were added to the electrolyte. Figure 9 shows the result. Again a "plateau" shape of curve is obtained. The same behavior was also observed for the 10% sugar solution.

In figures 6 and 9, it can be seen that the "plateau" begins at different ratios of copper to zinc. In figure 6, the ratio was approximately 0.6 while in figure 9 it can be estimated to be somewhat less than 0.3 (zinc content being at least 150 ppb and copper content, approximately 54 ppb). Had the ratios been the same, it would have been very easy to estimate the zinc content. But in the present case, it becomes much more difficult to interpret the phenomena. Apparently there is no simple solution to the problem of the determination of the zinc content. Hence in order to keep on the initial objective it was decided to focus efforts on the determination of copper, lead and cadmium.

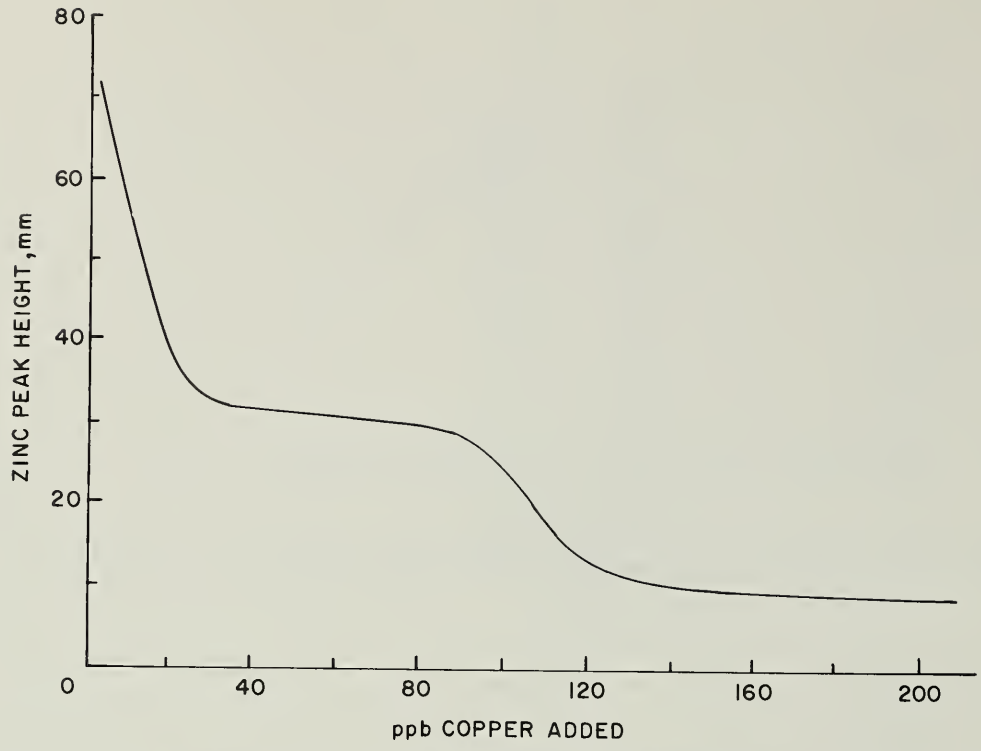

FIGURE 9--Zn peak height vs Cu added in presence of 150 ppb Zn.

The possibility of determining those species in refinery products other than refined white sugar was investigated. A molasses sample was studied and it was found that chloride ions, at a high concentration relative to the species considered, greatly affect the accuracy of the determinations made. The interference from chloride ions in electroanalytical analysis has been previously reported [1,8]. It has been observed that at high chloride concentrations an acid-soluble, nonstripping, brown layer forms onto the surface of the electrode. That phenomena was suspected to be of an electrochemical nature since it does not occur when the electric circuit is kept opened. This is the reason for avoiding any chloride-containing reagents, such as hydrochloric acid to adjust the pH of the electrolyte, or alkylchlorosilanes to prevent metal ions adsorption on the walls of the cell.

CONCLUSIONS

In conclusion, some heavy metals ions can be determined in sugar products using the described method of DPASV. Nevertheless, the technique has strong limitations. It appears that a dropping mercury electrode could best meet the requirements of the analytical chemist in the sugar industry where the determination of species like iron are of great importance. On the other hand, a rotated MFE allows the user to monitor very low levels of those ions that can readily be analyzed.

ACKNOWLEDGEMENT

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DISCUSSION

N. H. Smith(C&H): How does your method compare with atomic absorption for analysis of metals?

R. Cormier: This method allows you to quantitate some species with a high accuracy and for a much lower initial cost than that for atomic absorption (AA). A corresponding sensitivity with AA commands a price of $17,000 to $19,000 in Canada (say some $13,000 to $14,000 in U.S.); this includes a graphite furnace and a deuterium background corrector as accessories. In the case of DPASV, an investment of some $5,000 is required (up to $7,000 if the automatic controller accessory is used).

Interestingly, it is possible with this method to avoid sample pre-treatments, which is a definite asset over AA. Nevertheless, DPASV has limitations: hence, so far, only nickel, iron, cadmium, zinc and potentially, tin and arsenic species can be looked at with the demonstrated sensitivity. So, AA definitely offers a wider scope of analysis for metallic cations. But, on the other hand, one can quantitate some anionic species with the described instrumentation with a few simple modifications.

Finally it is not a question of which one is better than the other; they are complementary techniques for inorganic analysis.

N. H. Smith: Is it easy to train somebody to use it? Simplicity of operation is an advantage with atomic absorption.

R. Cormier: The answer is yes, if you have well-tried and well developed procedure on hand. The use of the electroanalytical automatic controller simplifies the work: just knowing which button to press and when to press it produces the required polarograms.

The chemist is then freed for interpretative work or special case studies.

C. Gagnon: The major advantage is that you don't have any sample pretreatment - you take the sugar, dissolve it and analyze it.

RAW SUGAR QUALITY ANALYSES, FROM 1968 TO 1976, AT THE NEW YORK SUGAR TRADE LABORATORY

By Walter Altenburg[1]

Since the early 1900's the New York Sugar Trade Laboratory (NYSTL) has filled the role of the unbiased third party in the measurement of polarization of sugars. Since 1968 the Laboratory has been testing the quality factors according to Contract No. 10 (now No. 12). The data presented here are those obtained since 1968. Filterability was formerly tested, but that is no longer one of the factors, so no data for filterability are shown.

Figure 1 shows the average value of polarization and the four quality factors for all sugars tested at NYSTL. This represents over 500 samples each year. These same data are also presented in table 1. Table 2 lists the distribution of premium and penalty cargoes for the factors: color and grain size. It

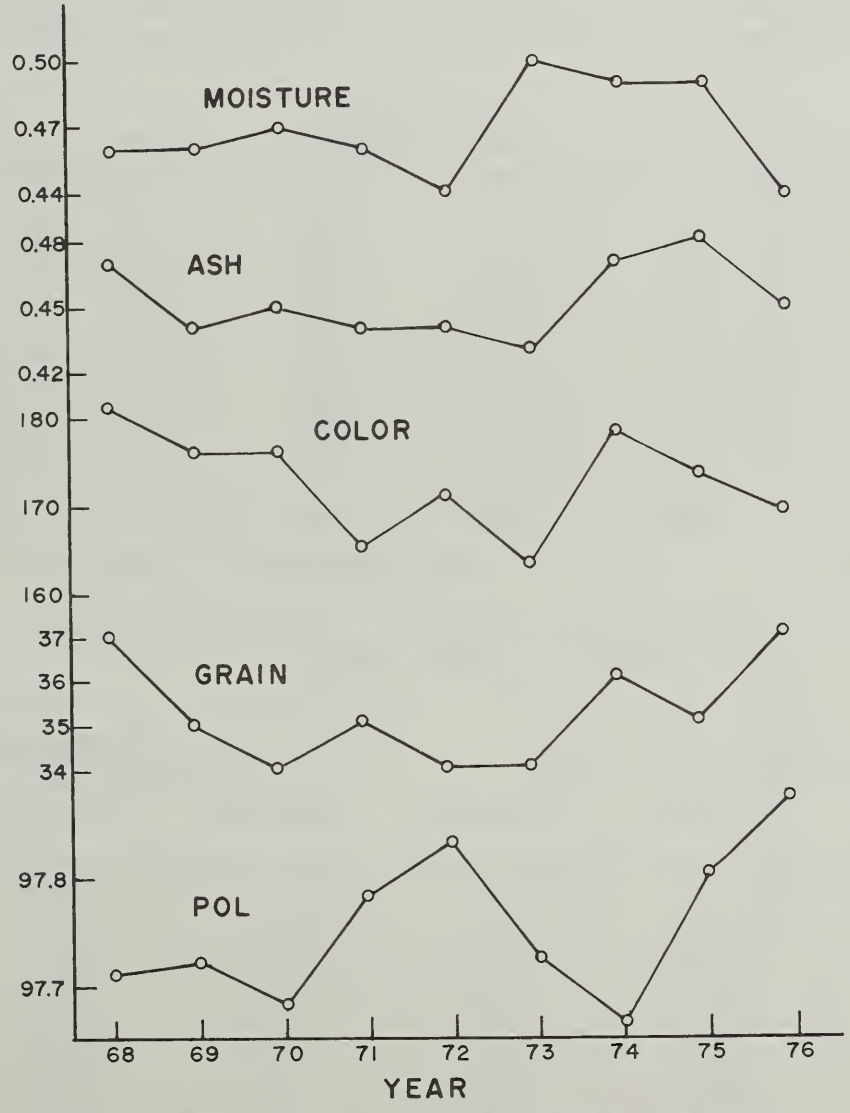

FIGURE 1--Average values for all sugars tested.

[1]President, New York Sugar Trade Laboratory, Inc., 300 Terminal Avenue West, Clark, NJ 07066.

should be noted that these are not world wide averages, but only NYSTL averages. Any changes or trends noted can partly reflect the changes in origins of sugars that came to the NYSTL. Table 3 shows the quality factors broken down by origin. Figures 2 through 8 show these data, but always in relation to the 1968 values. These plots, therefore, show how these origins have changed since 1968. Also shown are the distributions of premium and penalty cargoes for color and grain size.

These data are the historic facts, and may be examined from many different points of view. In summary it can only be said that there are different trends for different quality factors and different origins. In the average for all origins, there was in general an improvement from 1968 to 1973. In 1974 there was a worsening. In 1975 and 1976 there was again an improvement. So, the quality factors were effective in improving the raw sugar except in the unusual year of 1974 when there was a severe shortage and prices were high. In that year the emphasis was on more sugar, not on high quality.

TABLE 1--Average values of quality tests (all sugars tested)

	No. of samples	Moisture	Ash	Color	Grain	Polarization
1968	569	0.46	0.47	181	37	97.71
1969	501	0.46	0.44	176	35	97.72
1970	579	0.47	0.45	176	34	97.68
1971	562	0.46	0.44	165	35	97.78
1972	488	0.44	0.44	171	34	97.83
1973	561	0.50	0.43	163	34	97.72
1974	648	0.49	0.47	178	36	97.66
1975	483	0.49	0.48	173	35	97.80
1976	517	0.44	0.45	169	37	97.87

TABLE 2--Percentage of samples in premium, penalty and standard quality range

	Color (Standard range 100 - 230)			Grain size (Standard range 20 - 55)		
	Penalty	Standard	Premium	Penalty	Standard	Premium
1968	15.7	79.9	4.4	5.6	83.7	10.7
1969	16.4	76.3	7.3	6.2	83.7	10.8
1970	14.0	80.7	5.3	4.7	84.8	10.5
1971	9.1	84.2	6.8	4.4	83.3	12.3
1972	8.8	85.9	5.3	4.7	82.2	13.1
1973	9.2	82.6	8.2	4.1	78.2	17.7
1974	16.4	78.4	5.2	4.2	85.5	10.3
1975	15.7	77.4	6.8	6.0	84.3	9.7
1976	17.4	66.9	15.7	8.1	81.4	10.4

TABLE 3--Average values of quality tests

	No. of samples	Moisture	Ash	Color	Grain
		ORIGIN I			
1968	10	.25	.33	109	17
1969	7	.18	.37	124	20
1970	12	.25	.35	119	22
1971	14	.26	.33	133	22
1972	15	.25	.36	128	21
1973	19	.26	.37	112	20
1974	15	.23	.34	125	24
1975	24	.34	.33	129	23
1976	12	.25	.37	119	23
		ORIGIN II			
1968	42	.71	.46	174	39
1969	51	.74	.47	207	35
1970	46	.71	.45	192	33
1971	35	.52	.44	166	32
1972	42	.51	.42	197	24
1973	33	.42	.36	176	29
1974	52	.46	.41	170	29
1975	26	.58	.45	170	27
1976	--	---	---	---	--
		ORIGIN III			
1968	78	.45	.44	201	45
1969	64	.38	.35	174	47
1970	73	.38	.35	169	44
1971	77	.45	.43	163	44
1972	76	.43	.40	163	43
1973	105	.50	.42	156	41
1974	112	.50	.47	171	38
1975	100	.44	.43	177	37
1976	82	.43	.41	196	36

	No. of samples	Moisture	Ash	Color	Grain
		ORIGIN IV			
1968	1	.23	.47	100	20
1969	21	.21	.44	104	12
1970	--	---	---	---	--
1971	--	---	---	---	--
1972	--	---	---	---	--
1973	45	.26	.39	129	15
1974	7	.25	.47	170	25
1975	29	.24	.39	146	23
1976	60	.22	.39	80	19
		ORIGIN V			
1968	58	.56	.56	178	33
1969	34	.52	.51	187	32
1970	47	.54	.56	199	28
1971	48	.59	.53	165	39
1972	49	.59	.54	177	37
1973	35	.68	.50	187	34
1974	34	.54	.51	193	33
1975	6	.62	.58	221	35
1976	--	---	---	---	--
		ORIGIN VI			
1968	65	.50	.42	201	45
1969	65	.43	.41	196	42
1970	83	.45	.39	210	40
1971	89	.42	.38	204	37
1972	79	.41	.38	197	37
1973	119	.46	.39	179	39
1974	102	.41	.39	193	39
1975	30	.48	.45	249	41
1976	33	.44	.38	251	44
		ORIGIN VII			
1968	40	.44	.68	219	40
1969	25	.38	.72	231	38
1970	51	.41	.65	195	34
1971	40	.39	.64	196	33
1972	34	.34	.64	183	39
1973	35	.38	.71	194	43
1974	61	.34	.63	193	39
1975	29	.40	.65	173	38
1976	38	.38	.67	191	40

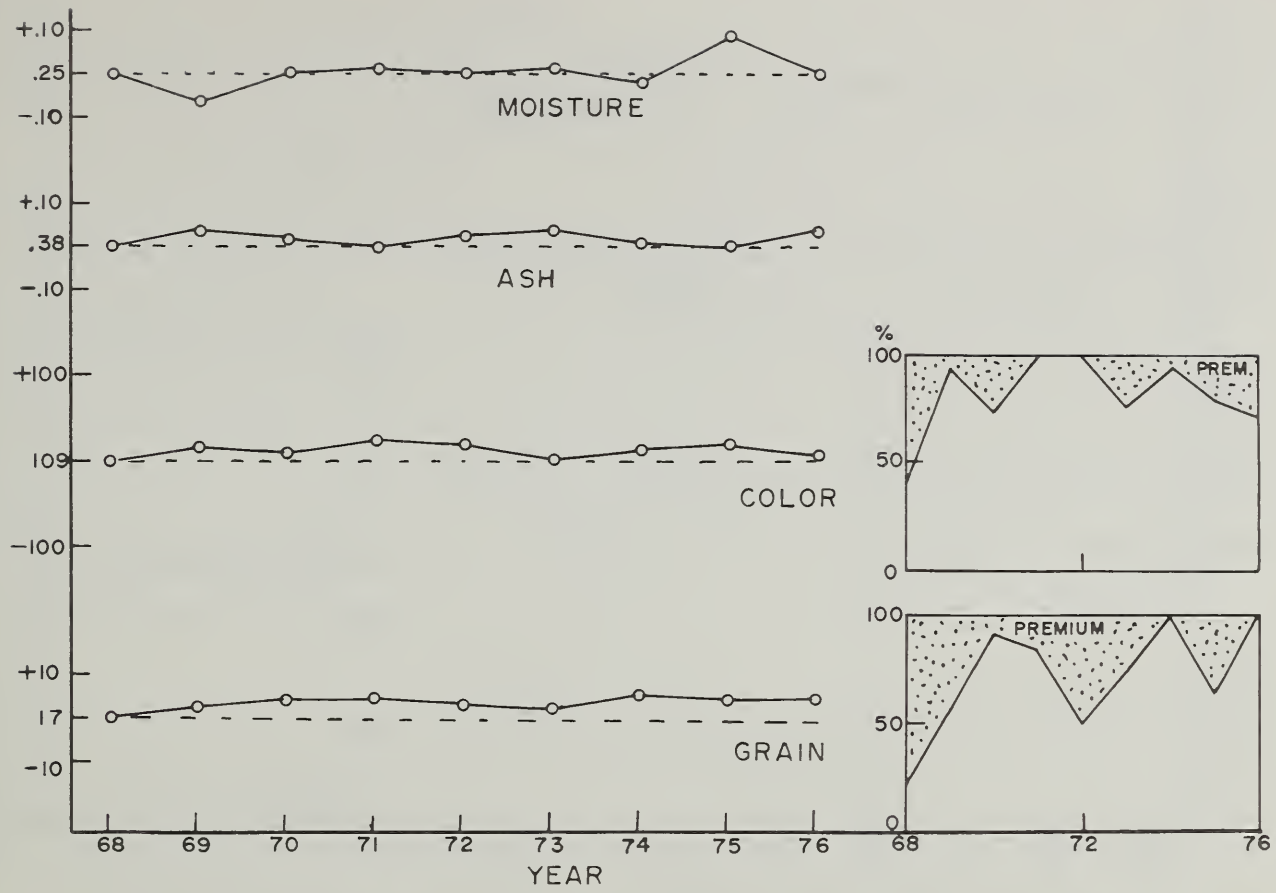

FIGURE 2--Change in quality factors since 1968 for origin I

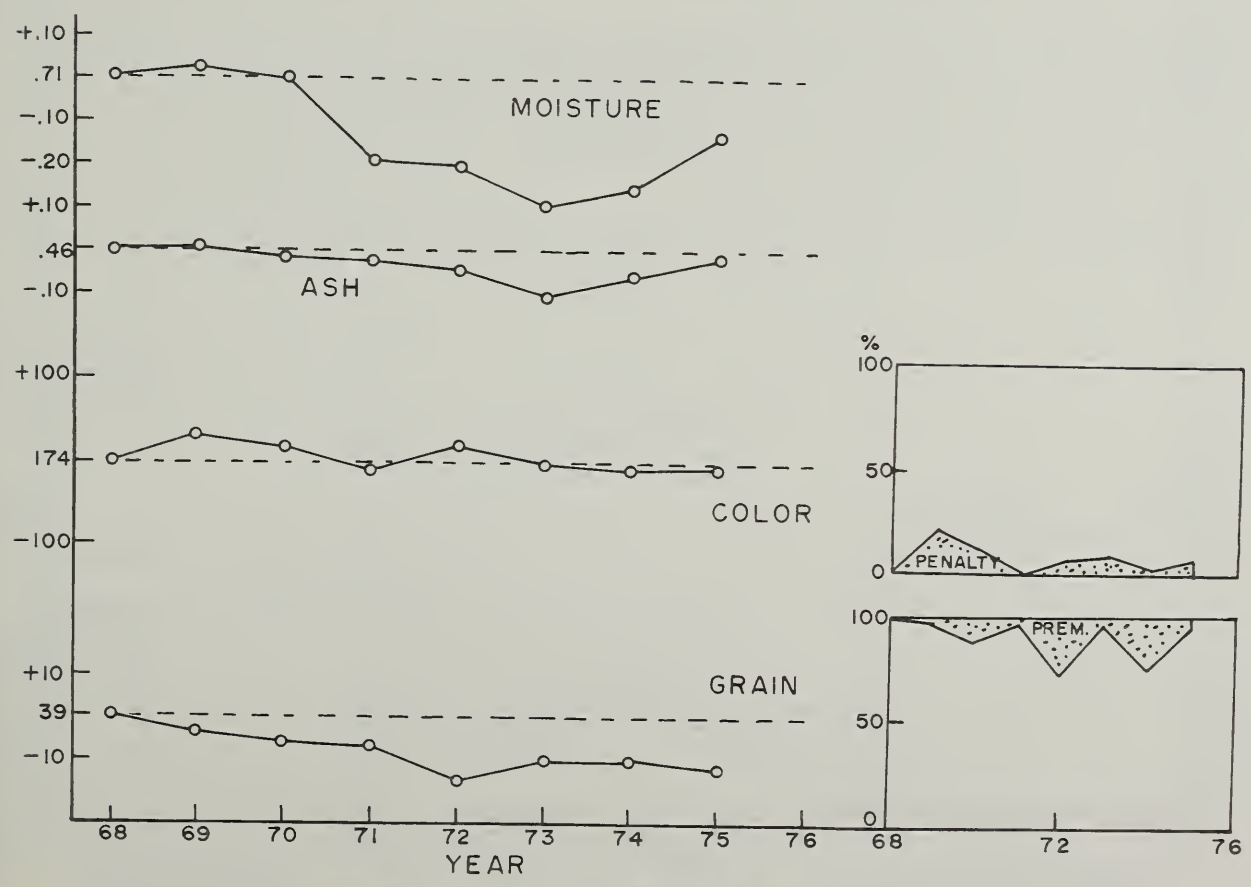

FIGURE 3--Change in quality factors since 1968 for origin II

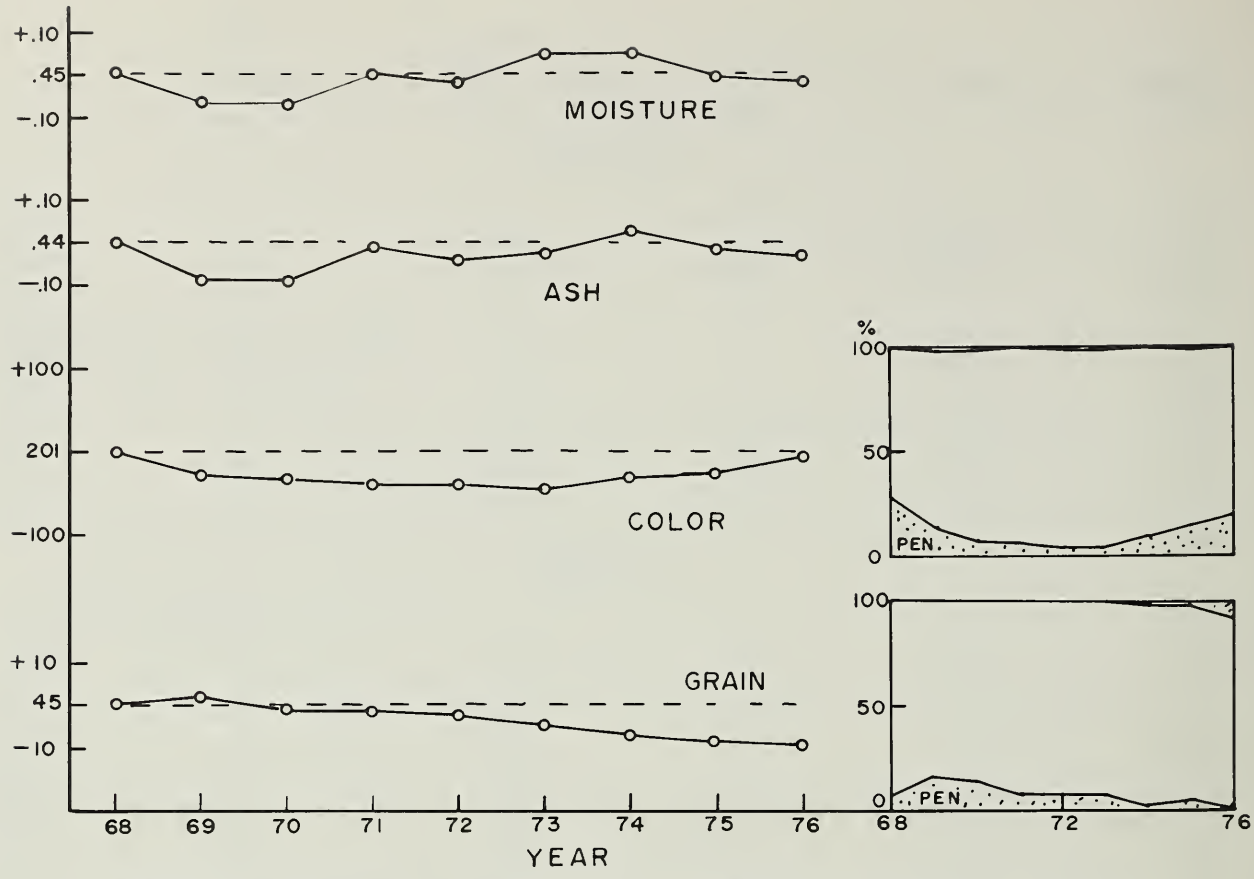

FIGURE 4--Change in quality factors since 1968 for origin III

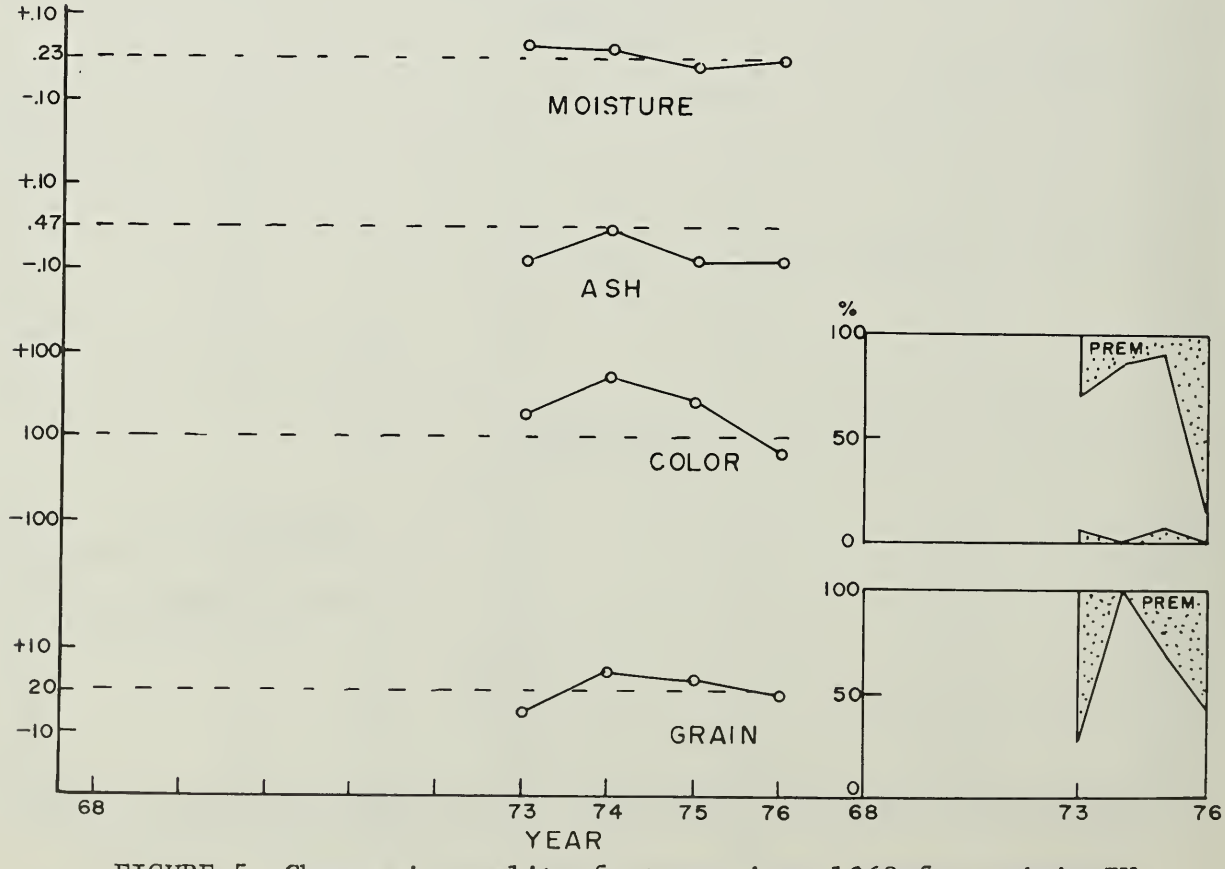

FIGURE 5--Change in quality factors since 1968 for origin IV

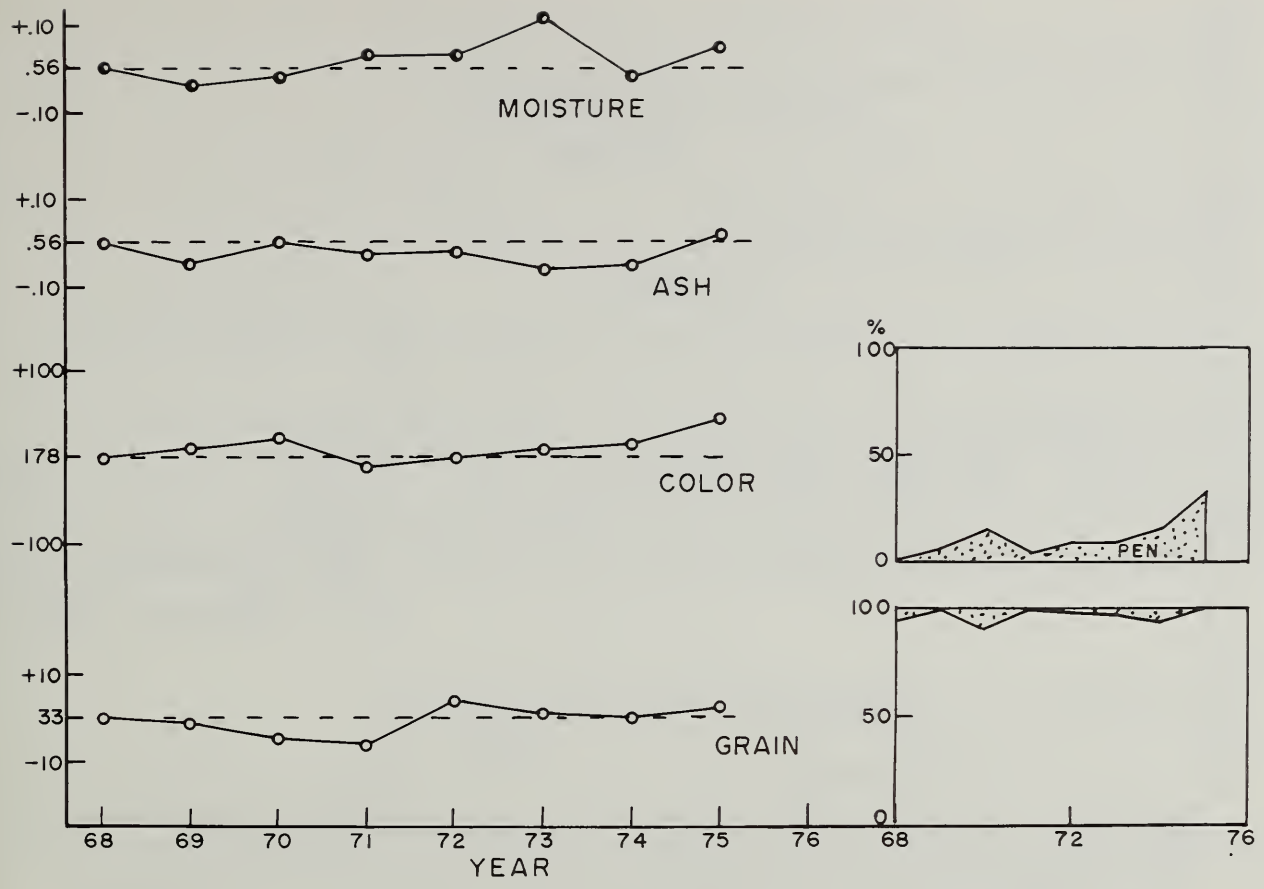

FIGURE 6--Change in quality factors since 1968 for origin V

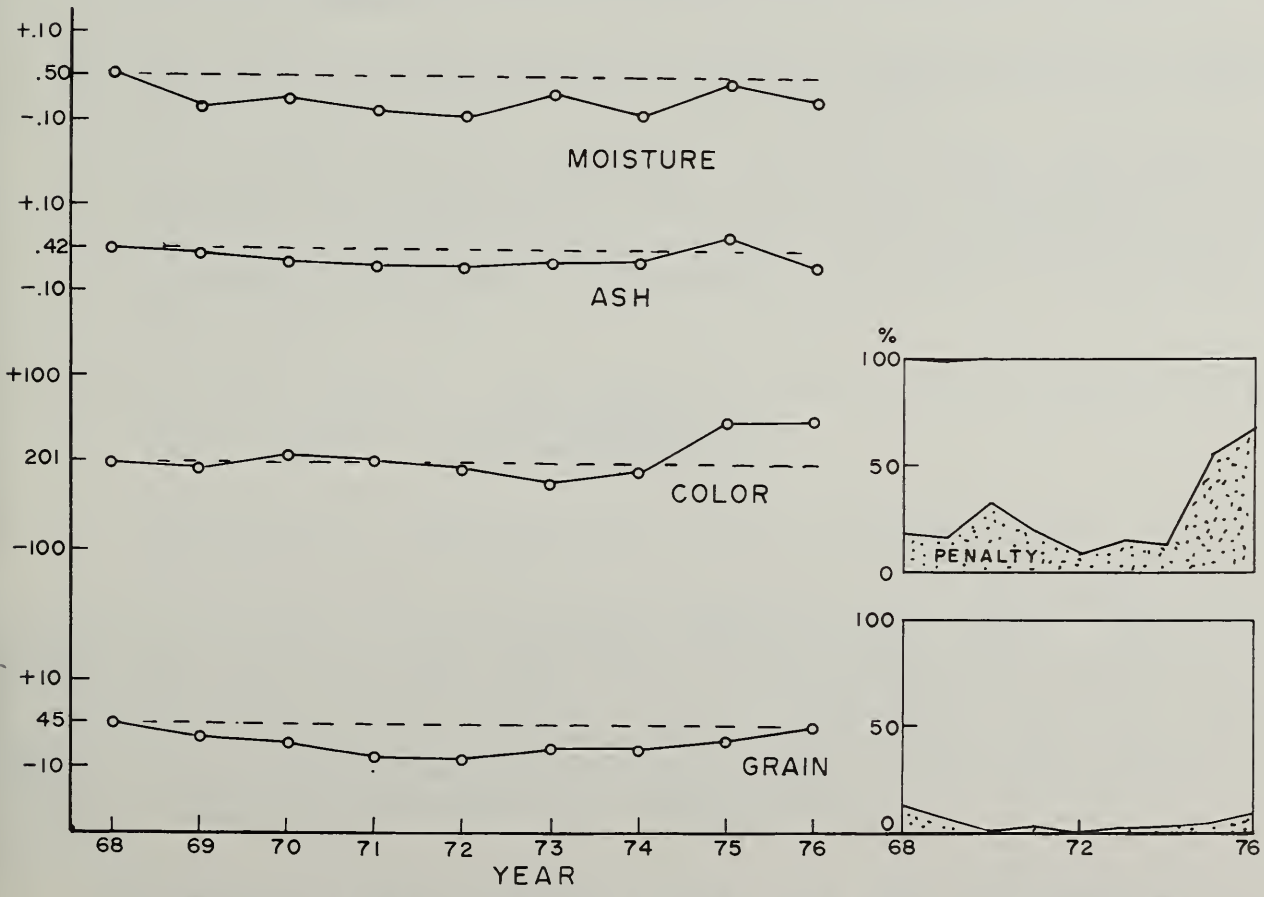

FIGURE 7--Change in quality factors since 1968 for origin VI

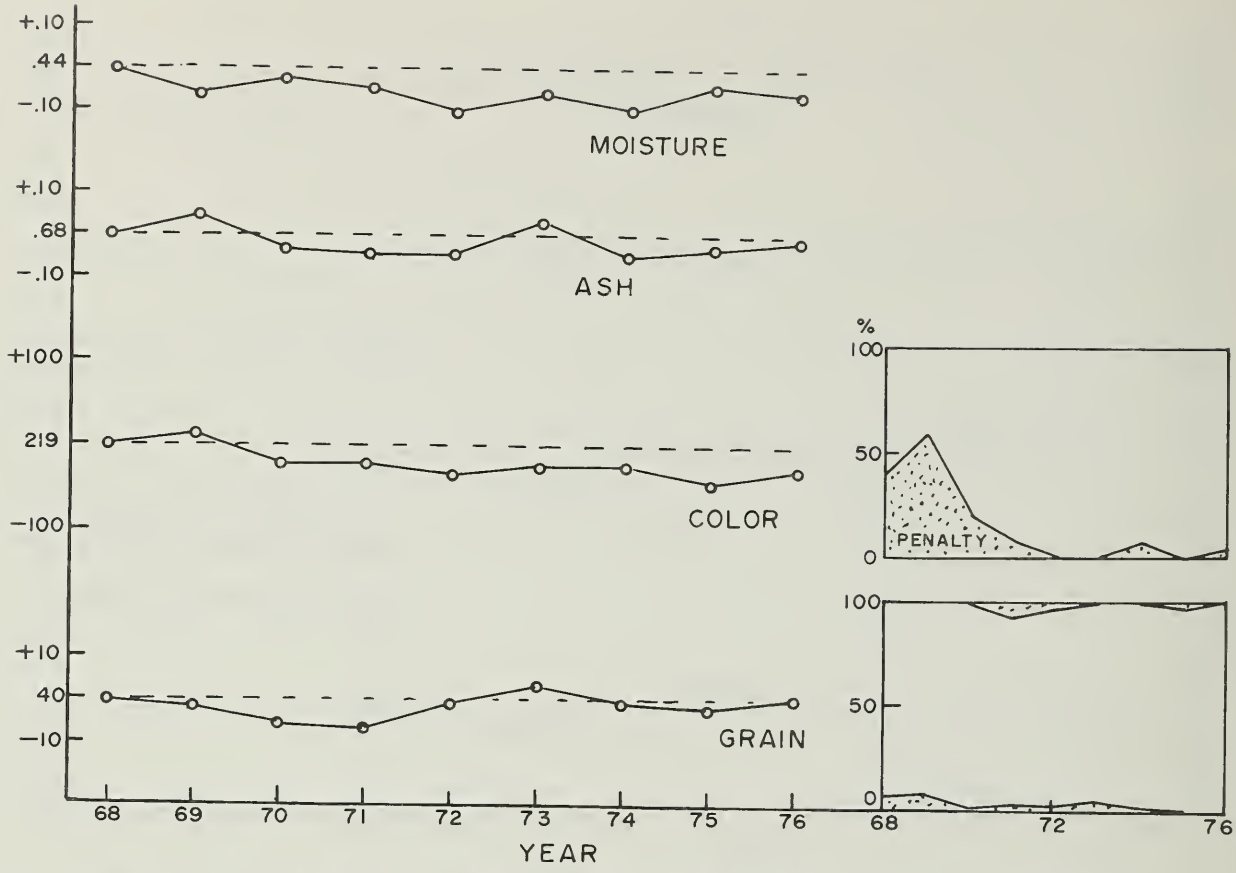

FIGURE 8--Change in quality factors since 1968 for origin VII

RAW SUGAR QUALITY CONSIDERATIONS AT SAVANNAH SUGAR REFINERY

By H. Richard Priester, Jr.[1]

INTRODUCTION

As do all refiners, we regard raw sugar quality as a vital concern.

In recent years approximately 90 percent of raws entering our process has been from South Florida. The other 10 percent has come from offshore sources such as Brazil, India, Thailand, and the Dominican Republic. Since this has been the case, characteristics of Florida raws have been our major considerations. Therefore, this presentation will give most attention to sugars we have received from that area.

CRITERIA FOR RAW SUGAR QUALITY

To satisfy our expectations we look for sugars with:

1. Characteristics consistent enough for uniformity of year round operations so as to assure minimum adjustments in process control.

2. Good and predictable filterability.

3. Polarizations sufficiently high enough to keep nonsugar load to our low grade recovery system from becoming excessive.

4. Low occluded ash in the crystal. Since the surface syrup film is separated from the crystal in the first stage of refining, it is hoped that the majority of impurity removal can be accomplished there, thereby allowing the remainder of the process opportunity to operate at peak efficiency.

5. A "Safety Factor" of less than 0.25 so as to avoid deterioration.
   $$\left(\text{Safety Factor} = \frac{\text{Moisture}}{100 - \text{Polarization}}\right)$$

6. Temperature of less than 100° F so as to prevent sucrose destruction.

We could easily see a 10 percent reduction in our avarage melt if we were to substitute raw sugars of quality lower than our normal supply. Filterability on various type raws has caused some refiners to decrease liquor Brixes to less than 60 to enable the material to filter. If raw sugars have excessively high temperatures or poor "safety factors", sucrose can be destroyed, and cost of operations would be increased by making necessary extra work in the process to remove additional impurities resulting from deterioration.

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[1]Chief Chemist, Savannah Sugar Refinery, P.O. Box 710, Savannah, GA 31402

It is evident that the initial condition of raw sugar we receive relates to dollars. Hence, the need for good and consistent quality cannot be over emphasized.

EVALUATION OF RAW SUGAR QUALITY

To evaluate raw sugar quality, samples are taken during unloading and while sugar is being introduced into the process. Routine analyses on these sugars include polarization, moisture, ash, invert, color, and pH (50.0 Brix solution). Values such as organic nonsugars, safety factors, and purities can be determined from these data. Daily tests are performed on process liquors taken from various stages of the refining operation. Should these fail to meet preset quality standards, additional checks may include further analyses on incoming raws.

SPECIFIC PROBLEMS RELATING TO RAW SUGAR QUALITY

Generally speaking, raw sugar quality at Savannah Sugar Refinery has a history of being good. However, we have not been totally free of problems. There have been occasions of poor filterability due to excessive insoluble material. This has caused reduced melts. There have been times when the occluded ash in the raw sugar crystal has been excessive, causing liquor from our first stage of refining to be above normal in ash. (We consider a normal raw sugar to have 70 to 80 percent of the ash in the surface syrup film.) When this has happened decolorization efficiencies of our carbonation station and char house have been decreased, making necessary various adjustments in process control.

Considering sugars received from South Florida, statements made previously concerning what we feel constitutes a good quality raw could be used in most instances to describe their characteristics. The worst problem we have encountered in sugars produced from that region has resulted from the high nitrogen content of their soil. This soil has offered the advantage of bringing about a fast-growing healthy plant, but the disadvantage is that amino acids from the ground are taken up in the cane and find their way into the finished raw sugar in small quantities. When the sugar has not been sufficiently cooled, the remaining heat becomes a catalyst that sets in motion a chemical reaction between these amino acids and the invert present. This reaction, called the Maillard Reaction, is exothermic (producing heat). As more heat is generated, the rate of reaction increases, and numerous negative effects are seen. The invert sugar breaks down into organic acids, thereby resulting in a drop in pH. Under these conditions sucrose is inverted, bringing about a loss in polarization. Also a marked increase in color of the sugar is seen. It then becomes necessary to increase the decolorization efforts in the refinery but without much effect as this color is most difficult to remove. The higher the temperature, the greater the color formation. In extreme cases where sugar was loaded at high temperatures, we have seen as much as a five degree drop in polarization in five days transport time from South Florida to Savannah. This has resulted in a loss of money to both the raw sugar producers and the refinery. Because of these occasional problems, efforts have been made by the Florida producers to assure proper cooling. Improvements have been seen, and such problems as just described are now rarely experienced.

The results of some of the studies made in our laboratory to determine effects of temperature on Florida raw sugar characteristics are given in tables 1 to 4 and figures 1 to 6. In these tests we held samples taken from various lots of Florida raw sugars in constant temperature ovens at various temperatures from 95°F to 125°F for a period of five weeks. In comparison, sugar was held at ambient conditions (80°F) for the same period of time. The containers were sealed to assure no moisture loss, and this was confirmed by laboratory analysis at the end of the five-week period. It was seen that at ambient conditions there was no change in analysis. However, as the temperature increased, polarization and pH significantly decreased, with color showing marked increases.

FUTURE EXPECTATIONS

Our future desires for raw sugar quality will be no different from our present criteria. We would hope that the raw sugar producers would make every effort to keep insoluble material to a minimum and storage temperatures well below 100°F. As always we would expect sugars with good filterability. With these standards of quality being honored we would anticipate no serious problems resulting from our incoming raw sugars.

TABLE 1--Raw sugar, lot "A"

Polarization	Moisture	Ash	Invert	Organic nonsugars	Safety factor	Purity	Color RBU	pH
98.60	0.28	0.47	0.34	0.31	0.20	98.88	24	6.1

Temperature, °F	80	95	110	125
Polarization	98.60	98.55	98.45	98.30
Color, RBU	24	38	54	104
pH	6.1	5.9	5.4	4.8

TABLE 2--Raw sugar, lot "B"

Polarization	Moisture	Ash	Invert	Organic nonsugars	Safety factor	Purity	Color RBU	pH
98.75	0.17	0.32	0.28	0.48	0.14	98.92	29	5.9

Temperature, °F	80	95	110	125
Polarization	98.75	98.70	98.60	98.50
Color, RBU	29	36	52	103
pH	5.9	5.7	5.3	4.9

TABLE 3--Raw sugar, lot "C"

Polarization	Moisture	Ash	Invert	Organic nonsugars	Safety factor	Purity	Color RBU	pH
98.85	0.21	0.40	0.24	0.30	0.18	99.06	30	5.9

Temperature, °F	80	95	110	125
Polarization	98.85	98.75	98.65	98.50
Color, RBU	30	42	57	106
pH	5.9	5.6	5.2	4.8

TABLE 4--Raw sugar, lot "D"

Polarization	Moisture	Ash	Invert	Organic nonsugars	Safety factor	Purity	Color RBU	pH
99.05	0.20	0.41	0.27	0.07	0.21	99.25	28	6.5

Temperature, °F	80	95	110	125
Polarization	99.05	99.00	98.95	98.85
Color, RBU	28	34	46	84
pH	6.5	6.2	5.8	5.1

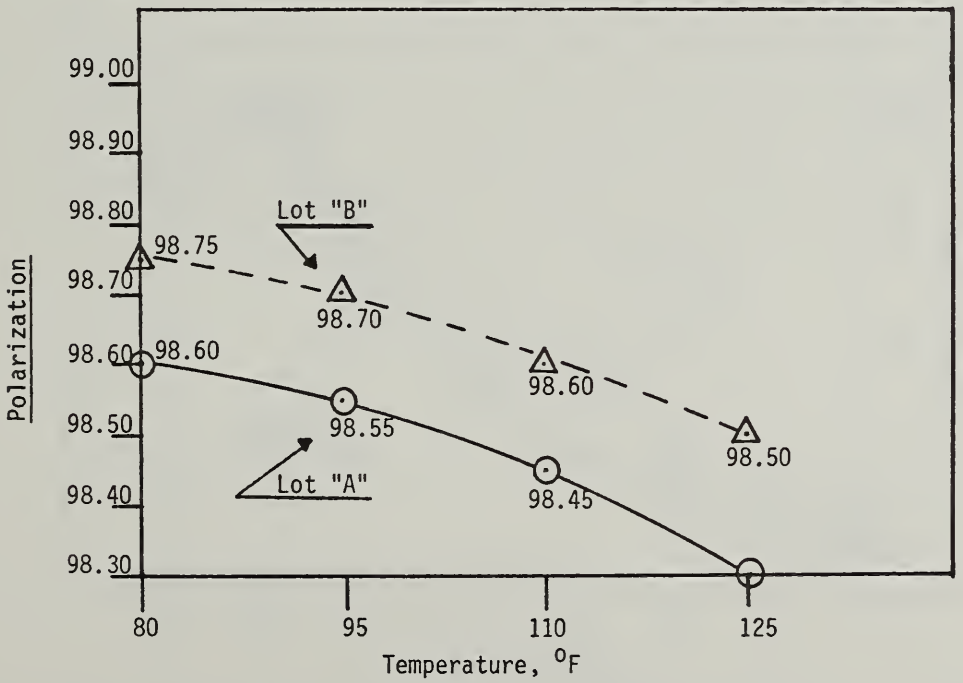

FIGURE 1--Effect of storage temperature on polarization for raw sugars.

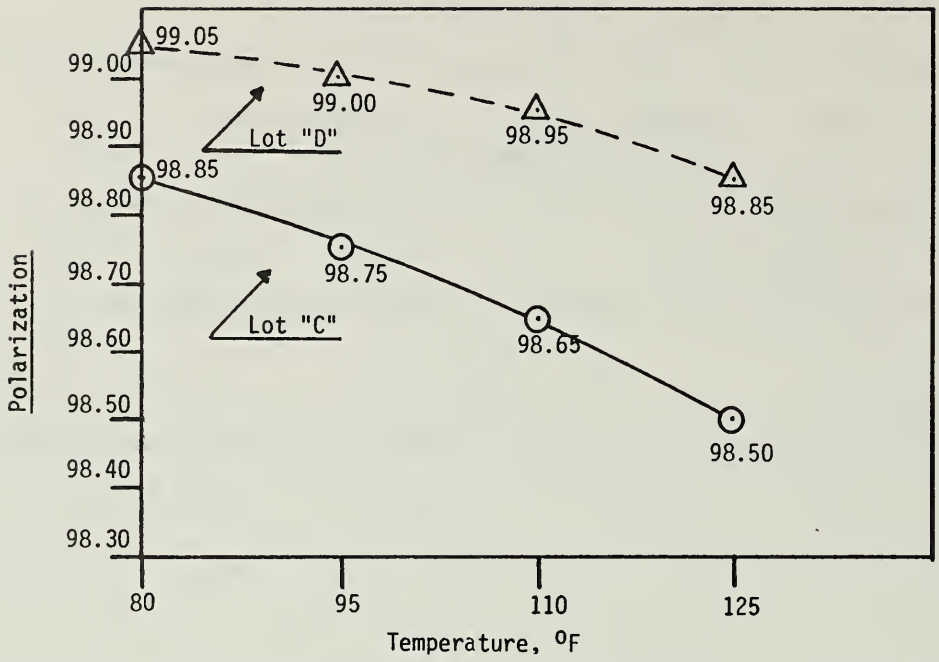

FIGURE 2--Effect of storage temperature on polarization for Florida raw sugars.

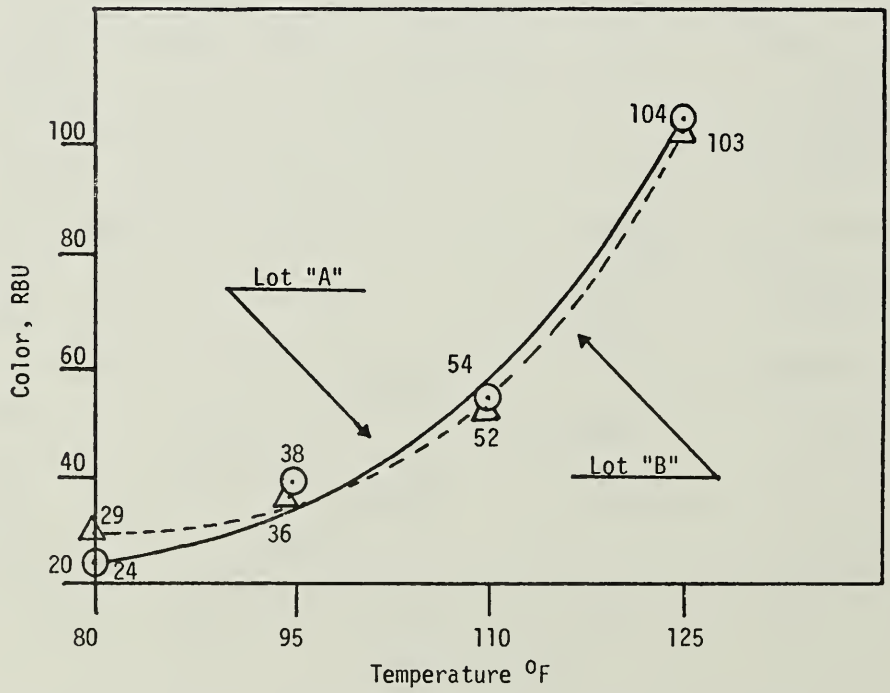

FIGURE 3--Effect of storage temperature on color for Florida raw sugars.

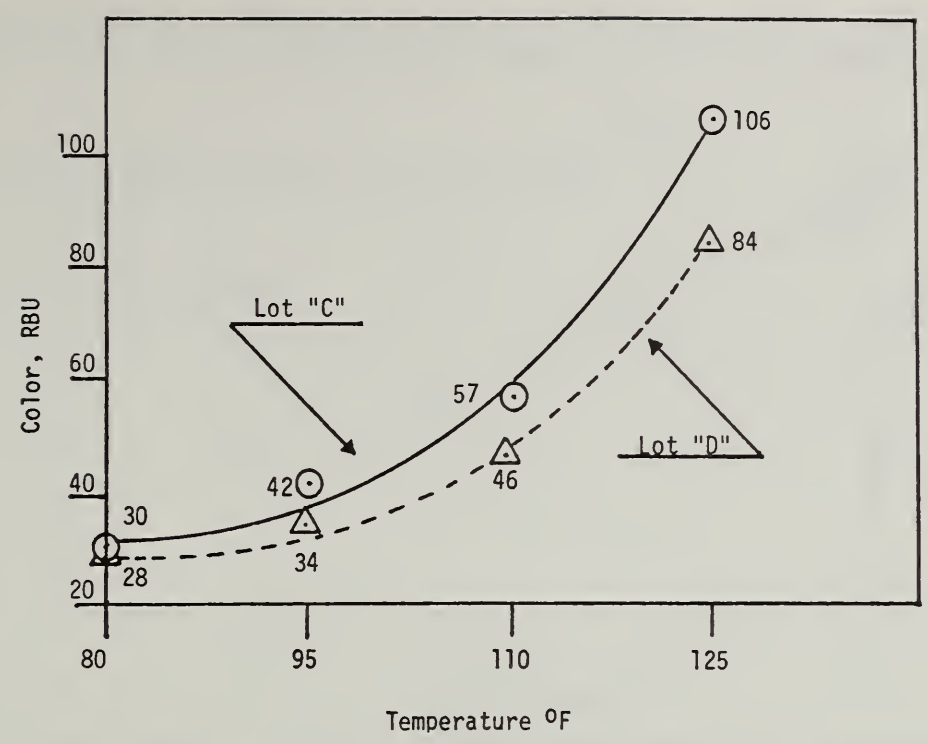

FIGURE 4--Effect of storage temperature on color for Florida raw sugars.

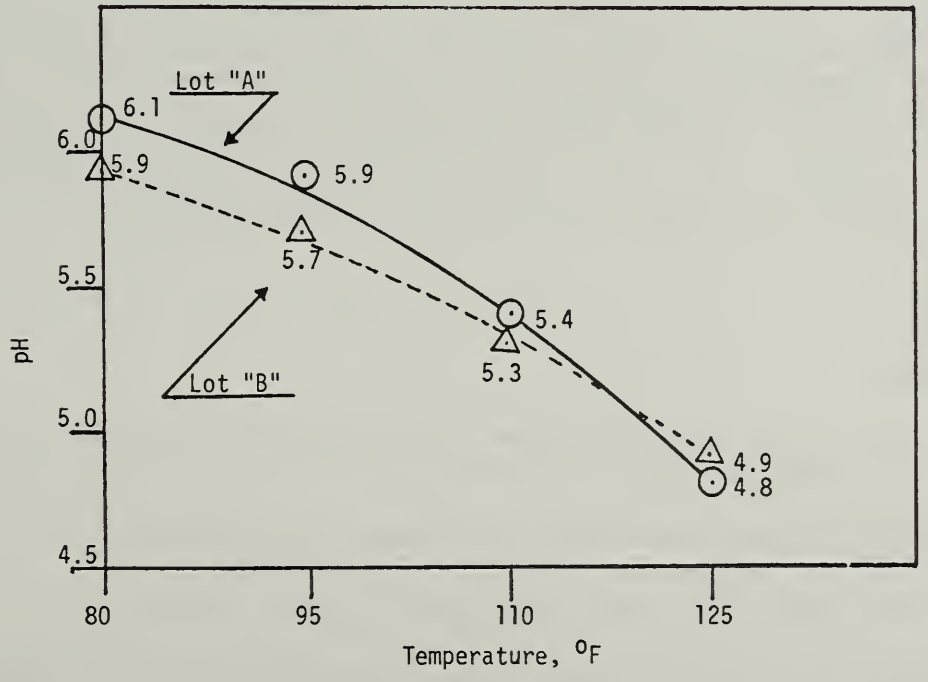

FIGURE 5--Effect of storage temperature on pH for Florida raw sugars.

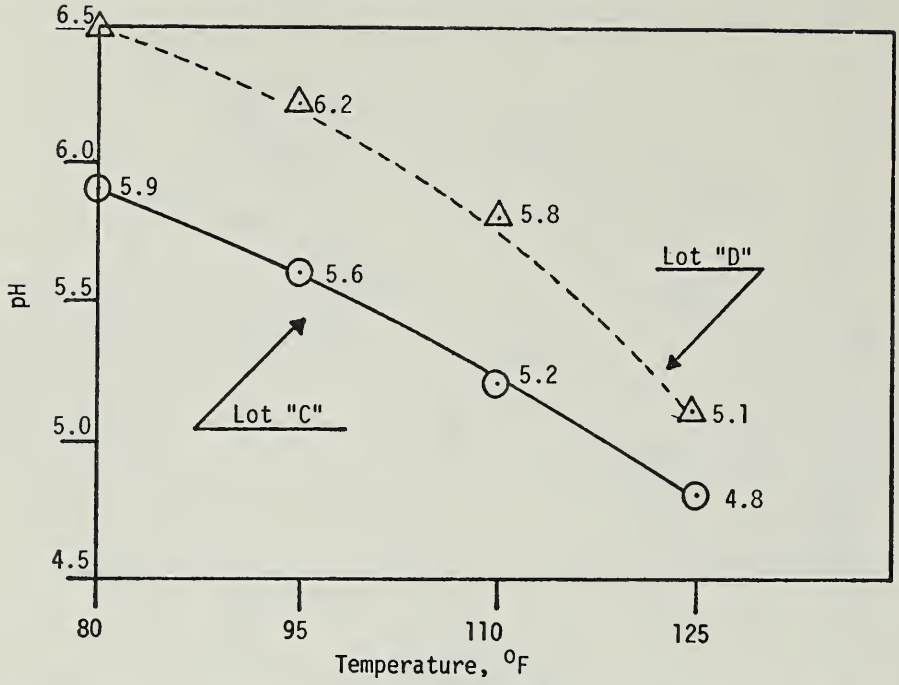

FIGURE 6--Effect of storage temperature on pH for Florida raw sugars.

DISCUSSION

M. Matic (SMRI): In speaking of filterability do you refer to filterability test results or filterability in the factory or filterability of carbonatated slurry?

H. R. Priester: What I'm speaking of in terms of filterability is how well the liquor goes through our Sweetland presses. The important factor is whether or not we have to shorten our filter cycles which of course results in a reduction in melt. If you're talking about filterability tests, you can show the same thing in the laboratory. But I am talking about filterabilty of carbonatated liquors. When we see filtration cycles on the press floor having to be reduced, and a resulting reduction in melt, we have poor filterability.

R. Moroz (Sucrest): Do you run nitrogen tests on the South Florida sugars?

H. R. Priester: No, we do not.

R. Moroz: Has South Florida solved the problem by removing the nitrogen compounds from the sugar or just by lowering the temperature of storage?

H. R. Priester: They have maintained lower temperatures.

R. Moroz: But they still have the same nitrogen content?

H. R. Priester: As far as I know, the nitrogen content has remained totally unchnaged. The soil characteristics in South Florida are basically the same now as they were when we experienced the color problems. However, since we do not routinely perform nitrogen analyses on our raw sugars, I cannot cite any comparative quantitative values.

L. C. Hayes (Amstar): Have you found correlation with the receipt of high temperature raws from Florida and degree of the quality of precipitate formed in carbonatation?

H. R. Priester: No, we see no relationship.

RAW SUGAR QUALITY STANDARDS

By Robert M. Gerstenkorn[1]

I have been asked to talk on the rather broad topic of raw sugar premiums and penalties from the point of view of the smaller refiner without bone char such as we have at Southdown.

We have had premiums and penalties now for nine years and we have seen some changes, but not any really significant changes, at least from our point of view, in the end product quality that have resulted from raw sugar changes.

Our case is primarily one dealing with ash removal. Our process employs phosphatation followed by granular carbon, ion-exchange, and then the standard evaporation, and a four-boiling system on the white side.

We are all faced with the same problem at times: a friend of the sugar refiners in Atlanta complains about ash, color, and other impurities. Primarily, we are faced with the problem of not being able to meet the ash specification with different raw sugars. So what we try to do is to take a look at the data on the incoming raw sugar and attempt to predict when we might run into an ash problem, or how we would handle the white side boilings. In doing this, we looked at the current ash criterion, which is ash as a percent of non-sugar solids, and we could find no correlation with ash in final product. The standard does tell us various things, but nothing useful from a control point of view, or from a cost point of view.

We then took a look at the ash in the whole raw sugar, and tried that correlation, and again saw no relationship. Figure 1 shows data from 1976. The points are two to two and a half month cutoff periods in which we looked at the ash in the input raw for that time against the percent of the input ash which wound up in the final three-sugar blend. For the whole raw ash, we see there is no correlation. However, a little farther along in the process, we found that the washed raw sugar ash gave a good correlation, and you get the result that you would expect; namely, that as the input ash goes up, the percent of that ash that appears in the refined product also goes up. It appears that we have a laundry with a fixed capacity and it can remove only just so much of any particular impurity. We decided that we could use these figures to predict what is going to happen.

In figure 2, we have a different plot showing the ash in both the two-sugar blend and the three-sugar blend. Here too we cannot see a correlation of final product ash with whole raw sugar ash, but there is an evident correlation with washed raw sugar ash. From these figures, we were able to tell the plant superintendent how, with a particular ash in his washed raw sugar, he would have to distribute his final product; that is, when he could go to certain blends, when he could go to packaging, and whether to increase or decrease liquid sugar production. In this way it became possible to move the whole product through the plant with a minimum of recycling. Improvement here will also obviously minimize costs.

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[1]Refinery manager, Southdown Sugars Inc., P.O. Bcx 1818, Houma, LA 70360.

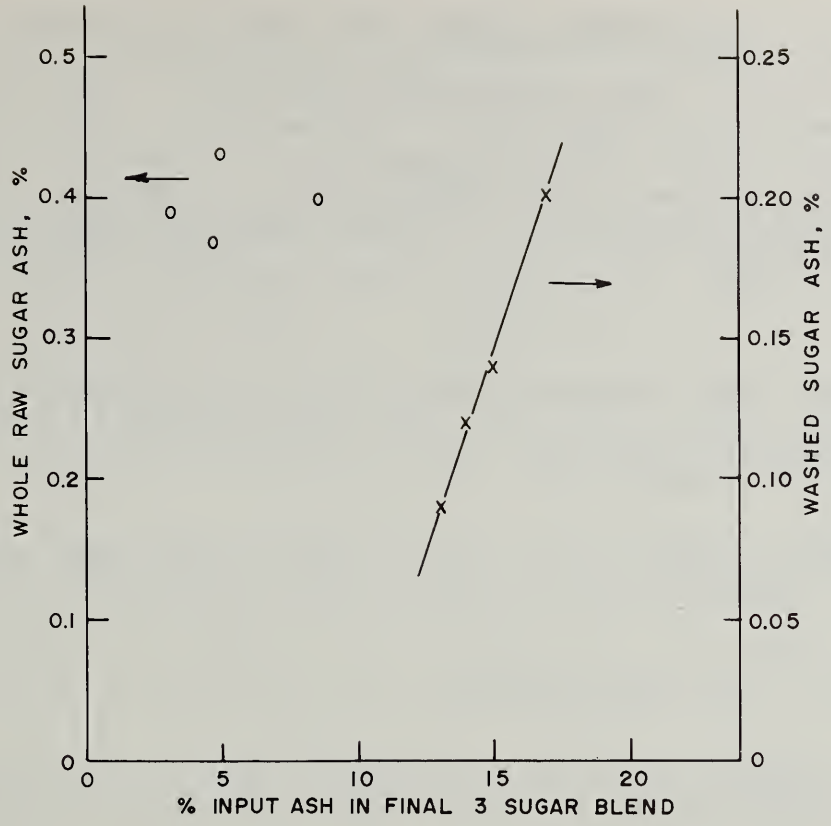

FIGURE 1-- Correlation of input ash with output ash in refined sugar.

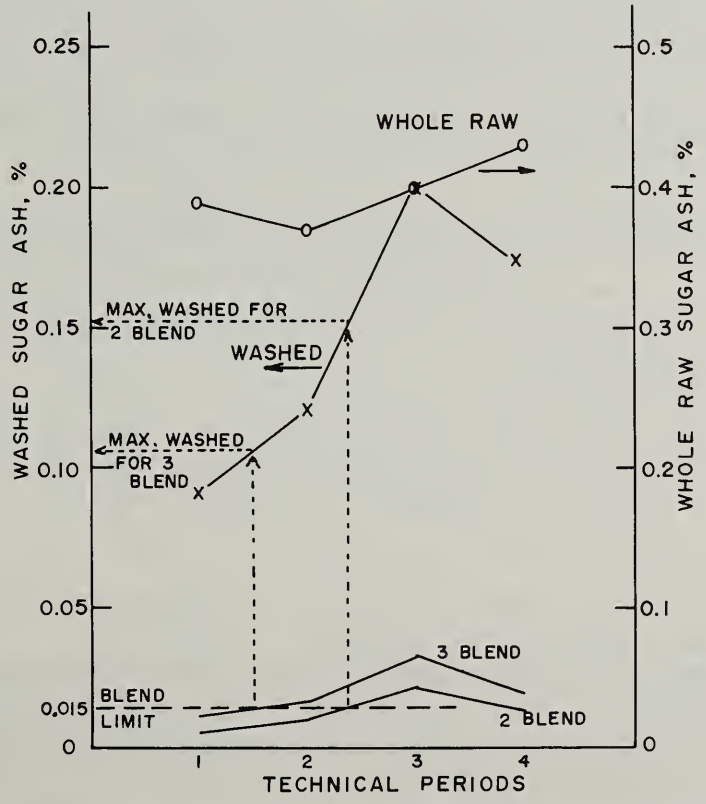

FIGURE 2-- Input and output ash for different technical periods.

Ash is the quality criterion which gives us the greatest problem. Each company has a different refinery: carbonatation or phosphatation or lime and filter aid; char, or no char; with ion exchange, or without. Each one of us has a capacity to remove certain impurities. I believe that we need to look at the ash penalty not only from the point of view of ash as percent of non-sugar solids, but also ash as a percent of the affined sugar. I believe that we need a two-pronged ash penalty: the existing one as is, plus an additional one.

DISCUSSION

M. Fowler (Amstar): How was ash determined? Is it sulfated or conductimetric ash?

R. M. Gerstenkorn: Conductimetric.

M. Fowler: Bottlers standard calls for a C ratio of 500 times the specific conductance. Have you found that your C ratio is different for your particular refinery?

R. M. Gerstenkorn: Not to my knowledge.

J. C. P. Chen (Southdown): Ash has been in my favorite subject for many years, since 1967 when contract No. 10 was introduced and now it has become effective internationally. It seems that for many years nobody brought out the importance of the ratio of ash to total nonsugar solids, and what that really means to our various refineries other than those of Amstar. In a recent review of this topic, I could see no interrelationship with the problem or the economical side of this ash/nonsugar solids ratio. In the mid-sixties there was a series of articles[1] presented by Dr. P. Honig and myself at SIT in which we tried to see what would be the real criterion of what this ratio will mean to the refinery. I commend Bob for finding out that one aspect is that the ash in the washed raw has a direct relation to the final product ash.

I believe this confirms some of our previous investigations, and I think that we will find the same relationship in the refinery. I am now on this refinery side, in contrast to when I was on the other factory side but we found the same results there. Ash is important for us as refiners from the chemical viewpoint and also on the economical side.

H. G. Gerstner: Dr. Chen was on the other side once when he was in Peru, and he received the Meade Award in 1967 for the best paper, mostly because it sounded like a brief by a defense attorney in a murder trial!

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[1] Honig, P., and Chen, J. C. P. 1964. Results of systematic quality control of Peruvian raw sugars. Proc. Sugar Ind. Technol. 23:130-145.

Chen, J. C. P., and Honig, P. 1964. The ash content of Peruvian raw sugars. Proc. Sugar Ind. Technol. 23:146-158.

Chen, J. C. P. 1967. Sulfated ash and total non-sugars in Peruvian raw sugars. Proc. Sugar Ind. Technol. 26:150-163.

RAW SUGAR QUALITY STANDARDS

By John V. Lopez Ona[1]

INTRODUCTION

The historical background of Raw Sugar Quality Standards is very extensive, and we cannot cover it all in a short presentation. Some of the highlights in the past nine years have been:

1. Evaluation of the proposed quality standards by the Raw Sugar Sellers Committee during several meetings which were held in 1966 and 1967. At those meetings, the proposed standards for ash, grain size, moisture or safety factor, filterability, and color were evaluated.

2. At the Twenty-Sixth Annual Meeting of the Sugar Industry Technologists in New York City, a technical paper was presented by Messrs. Culp and Hageney of American Sugar on "Raw Sugar Quality Standards."[2] The paper covered the proposed limits for moisture, or safety factor, and sulfated ash in whole raw sugar, and grain size, color, and filterability in affined raw sugar.

A "Symposium on Raw Sugar Standards" was also held during the above mentioned meeting[3] and a number of suggestions and recommendations were made by the attendees.

3. Effective January 15, 1968, the American Sugar Refining Company (now Amstar Corp.) included the Raw Sugar Quality Standards in their general contract provisions applicable to all contracts executed on "Bulk Raw Sugar Contract--Form 2021-65." For details on present Raw Sugar Quality Standards, see Appendix.

4. A second "Symposium on Raw Sugar Quality Standards" took place at the 1968 Technical Session of the Cane Sugar Refining Research Project in San Francisco[4]. Several representatives of U.S.A. refiners and one representative from Australia expressed opinions on the subject, while one representative from Peru sent his opinions by correspondence.

Today we are meeting here on a third "Symposium on Raw Sugar Quality Standards", nine years after the Contract No. 10 standards were put into effect (January 1968 to January 1977). The panelists have been asked to comment on:

1. Changes in quality since implementation of the standards.

2. Changes in emphasis on particular quality factors in their own operations.

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[1]Manager, Technical Services, National Sugar Refining Company, 1037 North Delaware Ave., Philadelphia, PA 19125.

[2]Culp, E. J., and Hageney, G. A. 1967. Raw sugar quality standards. Proc. Sugar Ind. Technol. 26: 164-173.

[3]Symposium, 1967. Raw sugar standards. Proc. Sugar Ind. Technol. 26: 174-177.

[4]Symposium, 1968. Raw sugar quality standards. Proc. Cane Sugar Refin. Res. 1968: 138-159.

3. Future trends in these directions.

CHANGES IN QUALITY SINCE THE IMPLEMENTATION OF THE STANDARDS

The National refinery in Philadelphia melts only offshore raw sugars from different parts of the world. The countries of origin during each calendar year of this study varied between 14 and 16 while the 1968 Sugar Act was in effect; and 4 of those countries supplied over 50% of our needs.

This pattern changed with new approaches introduced in the sugar market (expiration of the Sugar Act in December 1974, raw sugar contracts between producers and refiners, and producers' selling policies due to falling prices and severe weather conditions). While the number of countries of origin remained approximately the same, the four who had been main suppliers no longer provided us with the greatest share of our needs. This is shown in table 1.

The changes in quality since the implementation of the Raw Sugar Quality Standards are shown in trend charts for the four key countries above mentioned. The different parameters plotted are: (1) average raw sugar analyses per year for cargoes received by one refiner, shown in figures 1 to 5, and (2) average raw sugar quality standards per year for cargoes received by several refiners, shown in figures 6 to 9.

The trend charts are self-explanatory. Readers are invited to look over these trend charts, and draw their own conclusions. Damaged cargoes are excluded from these charts.

The Raw Sugar Quality Standards impose penalties and/or premiums on those raws outside the range of quality specifications. The amount of penalty or premium per pound for variances from the specifications is calculated according to

TABLE 1--Sources of raw sugar; % of incoming raws

Countries of origin	Old Sugar Market				New Sugar Market	
	1969	1970	....	1974	1975	1976
Brazil	10.02	11.26	....	17.27	7.52	0.0
Dom. Rep.	10.92	11.73	....	14.35	16.71	17.05
Peru	6.74	18.50	....	13.12	8.32	21.94
Philippines	28.67	28.88	....	29.91	10.71	0.0
	56.35	70.37	....	74.65	43.26	38.99
Others	43.65	29.63	....	25.35	56.74	61.01
	100.00	100.00		100.00	100.00	100.00

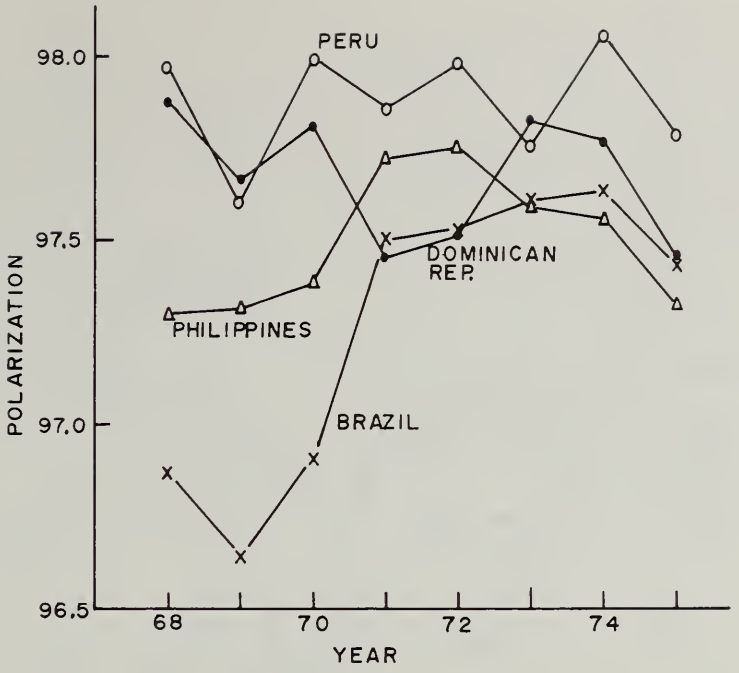


FIGURE 1--Average polarization of cargoes received at National.

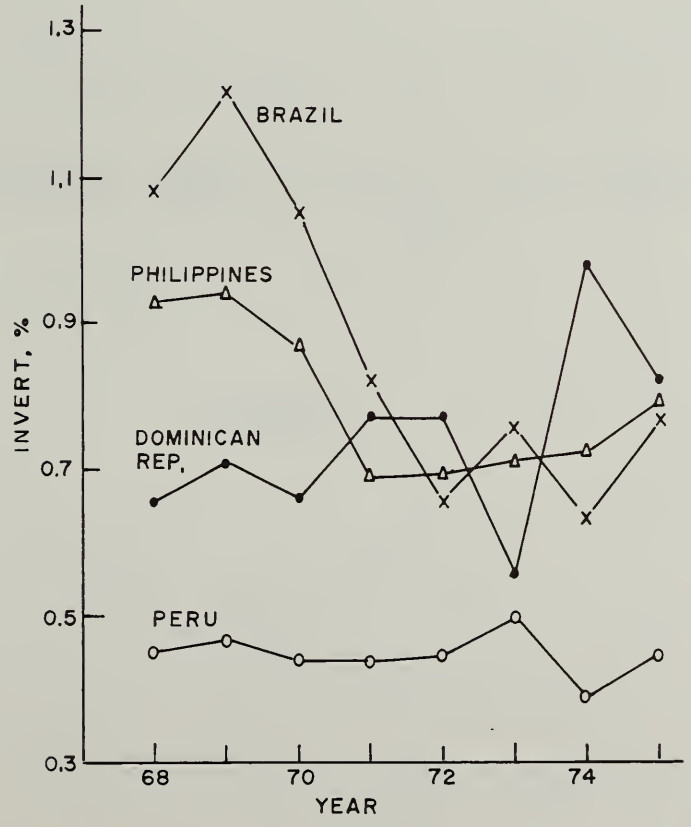


FIGURE 2--Average invert in cargoes received at National.

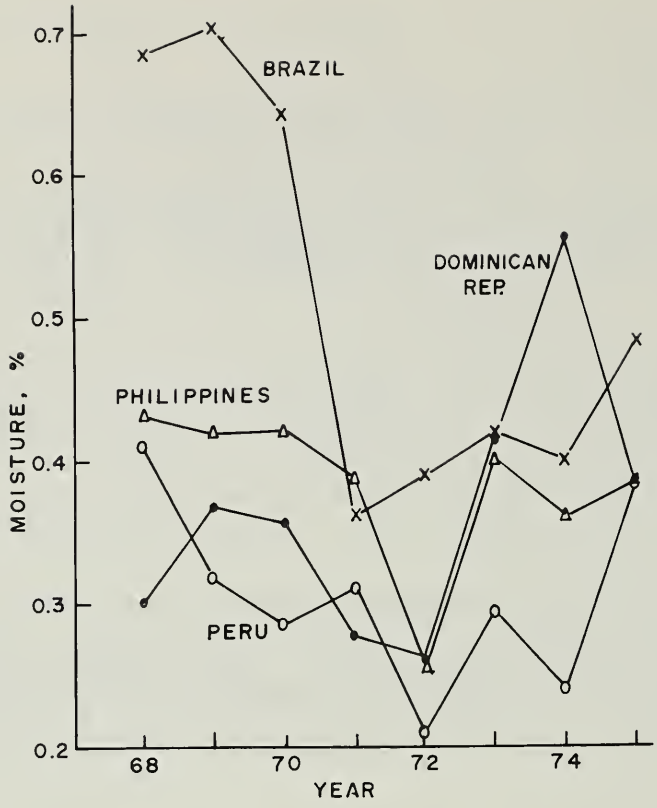


FIGURE 3--Average moisture in cargoes received at National.

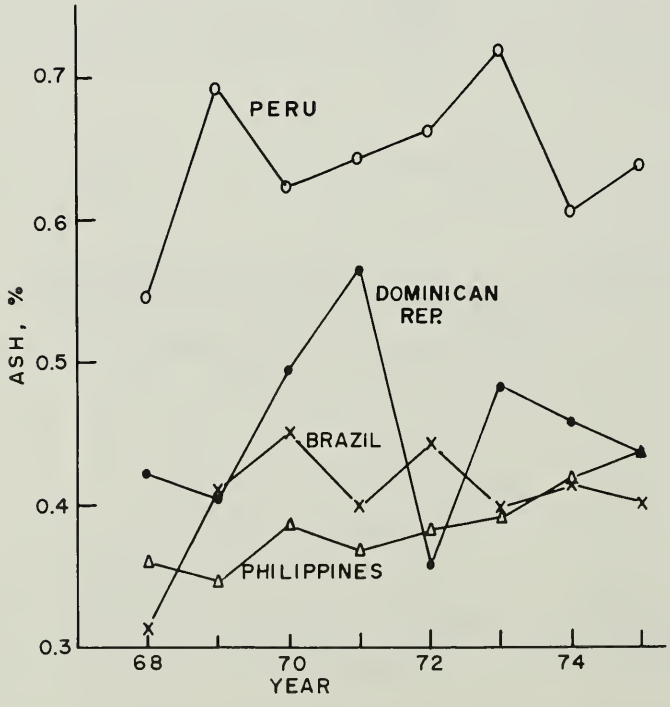


FIGURE 4--Average ash in cargoes received at National.

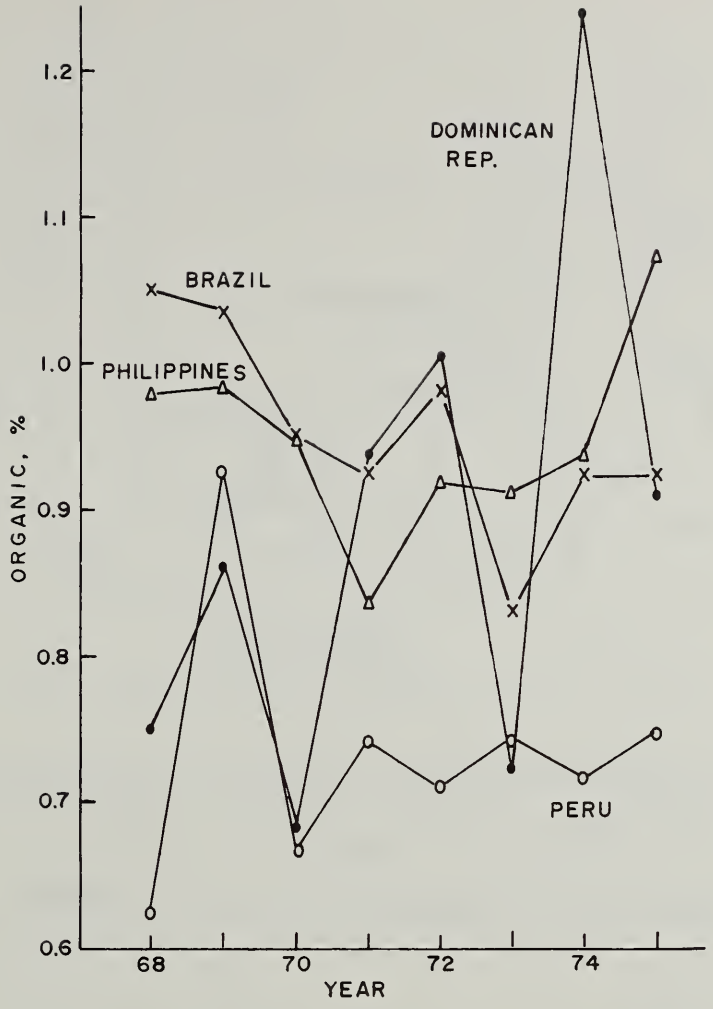


FIGURE 5--Average organic in cargoes received at National.

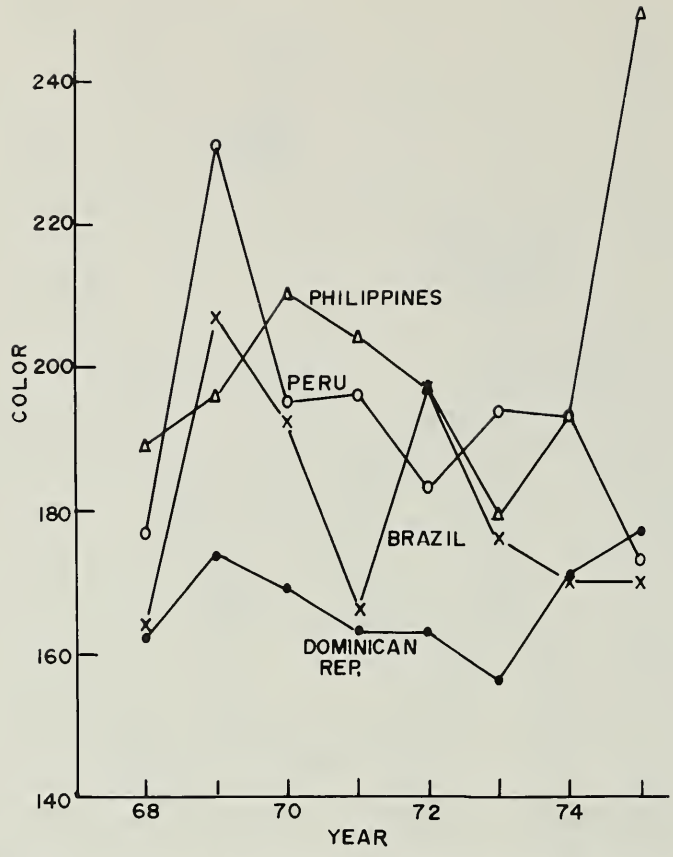


FIGURE 6--Average color of cargoes received by several refineries.

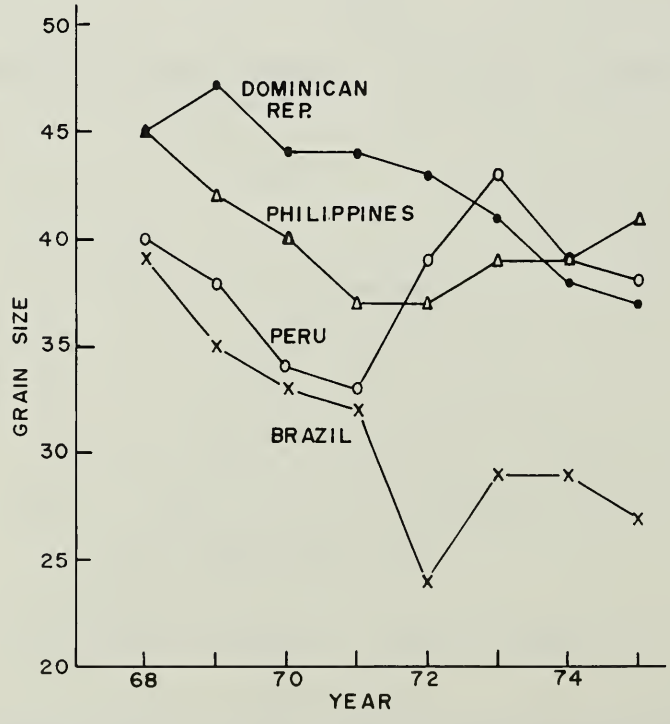


FIGURE 7--Average grain size of cargoes received by several refineries.

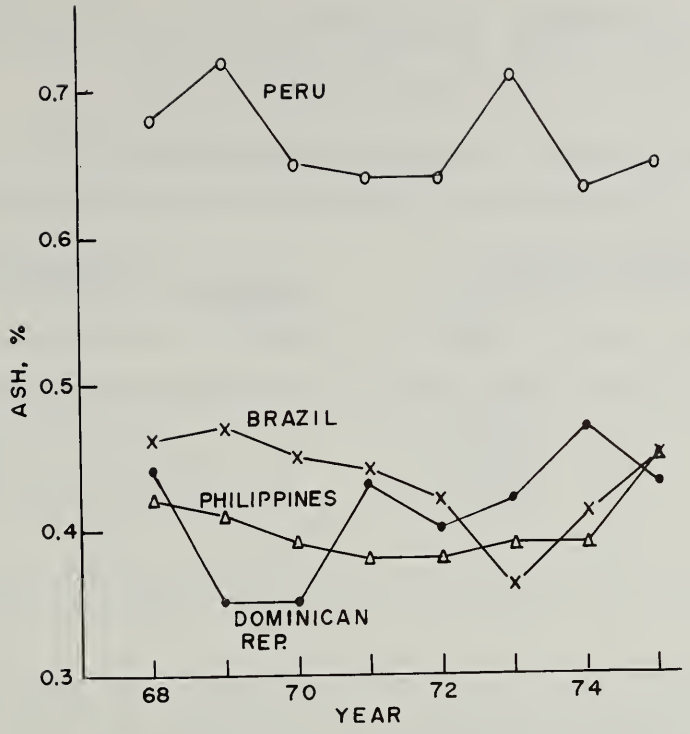


FIGURE 8--Average ash in cargoes received by several refineries.

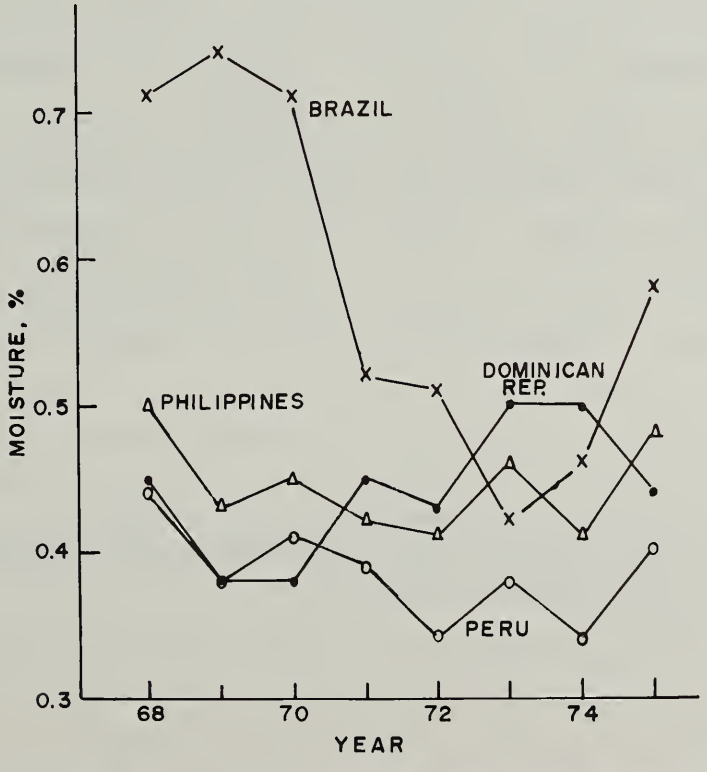


FIGURE 9--Average moisture in cargoes received by several refineries.

a table that establishes percentages of the basis price. The overall picture of premiums and penalties for the first and the last two years since the implementation of the quality standards can be summarized as follows:

TABLE 2--Frequencies of premiums and penalties

Years	Premiums				Penalties			
	Safety factor	Color	Ash	Grain size	Safety factor	Color	Ash	Grain size
1968	0	3	3	2	0	2	13	3
1974	0	0	1	3	0	8	7	1
1975	0	3	2	4	0	10	9	3

For the years of 1974 and 1975, the total frequency of penalties (16 and 22) significantly outnumber the total frequency of premiums (4 and 9). The penalties for color made the difference between the years compared. The significance is greater if presented in terms of "dollar effect" instead of "frequencies."

TABLE 3--Dollar effect of premiums and penalties

Years	Premiums				Penalties			
	Safety factor	Color	Ash	Grain size	Safety factor	Color	Ash	Grain size
1968	0	1,188	7,720	2,458	0	8,096	16,463	6,639
1974	0	0	2,030	18,167	0	55,342	15,599	40,259
1975	0	23,490	579	19,647	0	61,958	14,954	11,034

The details on the frequencies of premiums and penalties for 1974 and 1975 appear in tables 5 to 7, and the corresponding dollar effect in tables 8 and 9. Finally, in table 10, we have included the different costs involved in the testing of samples for Raw Sugar Quality Standards.

A summary of the dollar effect results for these two years can be used towards the evaluation of the economic effects of the system; as in table 4.

TABLE 4--Dollar effect of raw sugar quality standards

Raw Sugar Quality Standards	Dollar effect results 1974	1975
Gross penalties collected	$ 111,200	$ 87,946
Gross premiums paid	- 20,197	- 43,716
Net penalties collected	91,003	44,230
Cost of testing samples	- 4,965	- 5,642
To offset the economic effect of poor quality raws	$ 86,038	$ 38,588

In the specific case of poor quality raws, their effects resulted in an increase in the cost of production due to the loss of melt, decrease in sugar yields, usage of additional process materials, increased steam consumption, and so forth. To offset the economic effect of those raws, we collected $86,038 in 1974 and $38,588 in 1975. These figures cover a small part of the increase in the cost of production caused by the poor quality raws, and it would be interesting to know if anyone has had better experience than us on this matter.

In respect to the other goal of the Raw Sugar Quality Standards which was "to provide an incentive for sub-standard raw sugar producers to upgrade the quality of their finished product," the trend charts on figures 6 through 9 show that improvements were made by some countries on moisture and grain size. On the other hand, color has turned into a serious problem with certain raw sugars. During my trips in 1976 through South America, Central America, and the Caribbean visiting raw sugar producing areas supplying the U.S.A., I got the impression that :

1. some of the producers were not fully aware of the incentives to upgrade the quality of raws,

2. others felt that the incentives were not big enough to justify significant capital expenditures in process equipment, and finally,

3. most of them were more concerned with raw sugar prices than quality standards.

TABLE 5--Distribution of penalty and premium cargoes

	1974	1975
Penalty Cargoes	16	22
Premium Cargoes	4	9
Standard Cargoes	24	19
Total	44	50

Note:

Penalty cargoes are those which do not meet contract standards for safety factor, ash, grain size, and color; and, as such, the "Seller" is charged for the variation.

Premium cargoes are those which exceed the contract standards, and the "Buyer" pays extra for these quality sugars.

TABLE 6--Distribution of penalty and premium cargoes by country of origin (1974)

Country	No. of vessels	Premiums				Penalties			
		Safety factor	Color	Ash	Grain	Safety factor	Color	Ash	Grain
Peru	8						1	7	
Philippines	11						5		
Mauritius	1								
Dominican Republic	7			1			1		
Brazil	4				2		1		
Taiwan	1								
Jamaica	1								
Nicaragua	1								
Puerto Rico	2								
Swaziland	1				1				
Guatemala	2								
Trinidad	1								
British Honduras	1								
Australia	1								
Argentina	2								
Totals	44	0	0	1	3	0	8	7	1

SUMMARY

Total Cargoes	44
Total Premiums	4
Total Penalties	16

TABLE 7--Distribution of penalty and premium cargoes by country or origin (1975)

Country	No. of vessels	Premiums				Penalties			
		Safety factor	Color	Ash	Grain	Safety factor	Color	Ash	Grain
Argentina	4		2					3	1
Peru	5			1				5	
Malawi	1								1
Dominican Republic	12			1			1	1	
Jamaica	1								
Philippines	5			1			1		
Guyana	3						3		
Mexico	2								
Puerto Rico	2						2		1
Haiti	1						1		
British Honduras	1								
Australia	1								
Barbados	2						1		
Salvador	1								
Mauritius	1				1				
Brazil	3						1		
India	1		1		1				
Colombia	2								
Thailand	1								
Costa Rica	1				1				
Totals	50	0	3	2	4	0	10	9	3

SUMMARY

Total Cargoes - 50

Total Premiums - 9

Total Penalties - 22

TABLE 8--Dollar effect of premium and penalty cargoes by country of origin (1974)

Country	No. of vessels	Premiums				Penalties			
		Safety factor	Color	Ash	Grain	Safety factor	Color	Ash	Grain
Peru	8						2,609	15,599	
Philippines	11						38,471		
Mauritius	1								
Dominican Republic	7			2,030		7,174			
Brazil	4				13,260		7,088		
Taiwan	1								
Jamaica	1								
Nicaragua	1								
Puerto Rico	2								
Swaziland	1					4,907			
Guatemala	2								
Trinidad	1								
British Honduras	1								
Australia	1								
Argentina	2								40,259
Totals	44	0	0	2,030	18,167	0	55,342	15,599	20,259

SUMMARY

Total Premiums - $ 20,197

Total Penalties - 111,200

Net Difference (Penalties) $ 91,003

TABLE 9--Dollar effect of premium and penalty cargoes by country of origin (1975)

Country	No. of vessels	Premiums			Penalties			
		Safety factor	Color	Ash	Grain	Safety factor	Color	Ash	Grain
Argentina	4		4,677					3,762	6,848
Peru	5				167			9,873	
Malawi	1								1,469
Dominican Republic	12			114			350	1,319	
Jamaica	1								
Philippines	5			465			10,379		
Guyana	3						10,678		
Mexico	2								
Puerto Rico	2						26,780		2,717
Haiti	1						200		
British Honduras	1								
Australia	1								
Barbados	2						9,499		
Salvador	1								
Mauritius	1				1,938				
Brazil	3						1,072		
India	1		18,813		16,011				
Colombia	2								
Thailand	1								
Costa Rica	1								
Totals	50		23,490	579	19,647		61,958	14,954	11,034

SUMMARY

Total Premiums - $ 43,716

Total Penalty - 87,946

Net Difference (Penalties) $ 44,230

TABLE 10--Costs for testing of samples

		Cost to Buyer
Testing by the New York Sugar Trade Laboratory		
Moisture --	$11.00	
Ash --	12.00	
Color --	33.00	
Grain Size --	33.00	
Total	$89.00*	$ 44.50
*Half this cost is paid by the Seller		
Mailing of samples to Trade Lab. (Two one-gallon jars at a mailing cost of $3.50 per sample.)		7.00
Cost of gallon containers ($0.89 each)		1.78
Refinery cost for testing (moisture, ash, color, grain size)		58.00
Raw sugar cost (approximately 6 1/2 lbs/sample Average price $0.12 per pound		1.56
Total cost per cargo		$112.84
Total cost per year:		
1974 -- 44 cargoes x $112.84 =	$4,964.96	
1975 -- 50 cargoes x $112.84 =	$5,642.00	

CHANGES IN EMPHASIS ON PARTICULAR QUALITY FACTORS IN OUR OPERATIONS

When we started implementing the Raw Sugar Quality Standards in 1968, all four quality factors being tested were considered equal in importance. Based on the data collected during the last nine years, the quality factors can now be classified in the following priority order based on dollar effects:

Premiums	Penalties
1. Grain size	1. Color
2. Color	2. Grain size
3. Ash	3. Ash
4. Safety factor or moisture	4. Safety factor or moisture

The grain size is the factor that ranks number one in premiums paid while color occupies the top position on penalties collected. The penalties for color have increased significantly in 1974, 1975, and 1976 for sugars from those countries that are warehousing their raw sugar over long periods of time. The standards for color are 230 to 100, and a raw sugar over 275 usually creates serious problems at the refinery.

Ash continues to be a problem with raw sugars from one well-known origin.

The safety factor test had little effect on premiums or penalties during the last nine years, and it would be very interesting to know if ours is an isolated case or if other refiners have found just the opposite of our experience. Since we are spending approximately $1,000 per year on a test that is giving little dollar effect results, the question of its need is brought to the attention of the participants in the symposium.

FUTURE TRENDS IN RAW SUGAR QUALITY STANDARDS

The manufacturer of any commodity is interested in the quality of his raw material because of the close connection existing between this factor and the yields, production costs, quality and uniformity of his finished product. Although he may have the best factory equipment and manufacturing organization possible to assemble, if raw materials of suitable quality are not available it is unlikely that his product can be manufactured and marketed at a profit. For this reason, in most manufacturing industries the raw materials receive their share of attention, and receive it first because it is much easier to avoid trouble in this manner than to cure it after the materials have entered the process.

The foregoing, at first thought, may appear exaggerated when applied to the sugar refining industry; but it is believed that a cargo of unsatisfactory raws every now and then will convince the most skeptical refiner. It is obvious from the start that if the refiner desires better raws, this must be made possible either by offering the raw sugar producer some extra financial inducement for sugars grading above a certain fixed standard with penalties attached to sub-standard sugars; by convincing the raw sugar producer that the quality of his product reflects certain manufacturing conditions existing in his plant which, if corrected, would yield him additional income from the sale of raws, or, as a last resort, by direct discrimination against the inferior product of certain origins. Market conditions would, of course, determine the effectiveness of the latter means; but, in any case the refiner would prefer not to employ this unless other methods prove ineffective.

After writing and talking about the refining qualities of raw sugars for more than fifty years, the refiners in the U.S. A. finally made a move in 1968 when raw sugar quality standards were included in the bulk contracts. To motivate the producers towards the improvement of the quality of their raws, the refiners offered financial inducement for raw sugars grading above a certain fixed standard with penalties attached to sub-standard sugars.

It is my personal opinion that the next step to take in the future is the "feedback" to all interested on the results obtained in the first decade since the implementation of the Raw Sugar Quality Standards. A Raw Sugar Buyers Committee should be formed to collect, analyze, and compare the results of the quality tests against the standards on an individual and group basis. This first "blue book" should then be distributed among the following entities:

1. Buyers of raw sugars so they can understand better the quality problems of each raw sugar origin.

2. Sellers of raw sugars in order to make them aware of their strong and weak points by an actual comparison of their performance against the standards and against other producers. At the present time, R. Markey & Sons is distributing the information in table 11 among various raw sugar suppliers.

3. United States Cane Sugar Refiners' Association in Washington, D. C. for distribution among government agencies, scientific institutions, universities, and so forth. I am sure that this feedback on Raw Sugar Quality Standards could be an important point to consider in any future legislation.

4. The New York Coffee and Sugar Exchange for distribution among sugar brokers throughout the world.

5. Others.

I am sure that all of us will profit from the exchange of ideas and opinions that will follow this panel's presentation.

TABLE 11--Frequencies and dollar effect of premiums and penalties.
Source: R. Markey * Sons, Inc., 99 Wall St., New York, NY 10005

Frequencies

Year	Cargoes sampled	Premiums				Penalties			
		Safety factor	Color	Ash	Grain size	Safety factor	Color	Ash	Grain size
1968	481	0	29	20	49	7	66	78	21
1969	437	0	28	41	49	9	75	44	19
1970	529	0	25	46	67	5	75	64	19
1971	482	0	30	23	73	9	58	68	22
1972									
1973									
1974									
1975	115	0	9	2	12	7	30	8	10

Dollar effect

Year	Cargoes sampled	Premiums				Penalties			
		Safety factor	Color	Ash	Grain size	Safety factor	Color	Ash	Grain size
1968	481	0	30,565	12,159	48,676	2,977	268,904	91,423	53,500
1969	437	0	20,595	13,887	56,113	14,161	503,075	66,608	53,973
1970	529	0	16,312	11,291	84,459	4,952	490,062	78,450	41,942
1971	482	0	19,904	4,239	96,374	5,667	282,254	71,337	53,861
1972									
1973									
1974									
1975	115	0	42,927	98	54,142	29,167	370,988	14,628	63,128

Appendix--Raw sugar quality system

For testing for refining quality, three one-gallon containers are to be filled completely, avoiding any headspace, the covers tightly affixed and one container forwarded to the Seller's Laboratory, one to the Buyer's Laboratory, and one to the New York Sugar Trade Laboratory. Each laboratory shall follow the procedures specified herein, and shall use the equipment and methods described for testing the whole raw sugar for moisture and soluble ash and for testing affined raw sugar prepared from teh whole raw sugar for grain size and color. Raw sugar within the following range of quality specifications will be considered sugar of standard quality.

	Standard quality range
Moisture	
Factor of safety (High number indicates low quality)	Not exceeding 0.30
Ash	
Ash content (percent of raw sugar)	Maximum standard ash content is derived by multiplying percent non-sucrose solids by the factor listed below which corresponds to the final polarization of the cargo:
	Up to and including 98.0° — 0.32
	Over 98.0° up to and including 98.2° — 0.33
	Over 98.2° up to and including 98.4° — 0.34
	Over 98.4° up to and including 98.6° — 0.35
	Over 98.6° up to and including 98.8° — 0.36
	Minimum standard ash content is derived by multiplying percent non-sucrose solids by — 0.16
Grain size	
Percent through 28 mesh Tyler (30 mesh U.S.) sieve (High number indicates low quality)	Between 55 and 20
Color	
I.C.U.M.S.A. Color Units Method 2 (1958) Modified (High number indicates low quality)	Between 230 and 100

The average of the two nearest of the three Laboratory test results shall be used as the basis for determining Refining quality under the Standard Quality Range unless two are equidistant from the median, then the median shall be used. Such average shall be determined separately for each quality determination.

The percentage penalty or premium per pound, for variances from the specifications set forth shall be determined pursuant to the following table, it being understood that penalties and premiums for variances from standard quality shall be applied separately for each specification and that fractions shall be in proportion.

Specification	
Moisture	
Factor of safety	For each 0.01 in excess of 0.30, deduct 0.06% of basis price
Ash	
Ash content (percent of raw sugar)	For each 0.01% of ash content in excess of derived maximum standard ash content deduct 0.01% of basis price; for each 0.01% of ash content below derived minimum standard ash content, add 0.005% of basis price.
Grain size	
Percent through 28 mesh Tyler (30 mesh U.S.) sieve	For each 1% above 55% deduct 0.04% of basis price: for each 1% below 20% add 0.02% of basis price.
Color	
I.C.U.M.S.A. color units Method 2 (1958)	For each unit above 230 deduct 0.01% of basis price: for each unit below 100 add 0.005% of basis price.

TEN YEAR TREND IN RAW SUGAR ANALYSIS

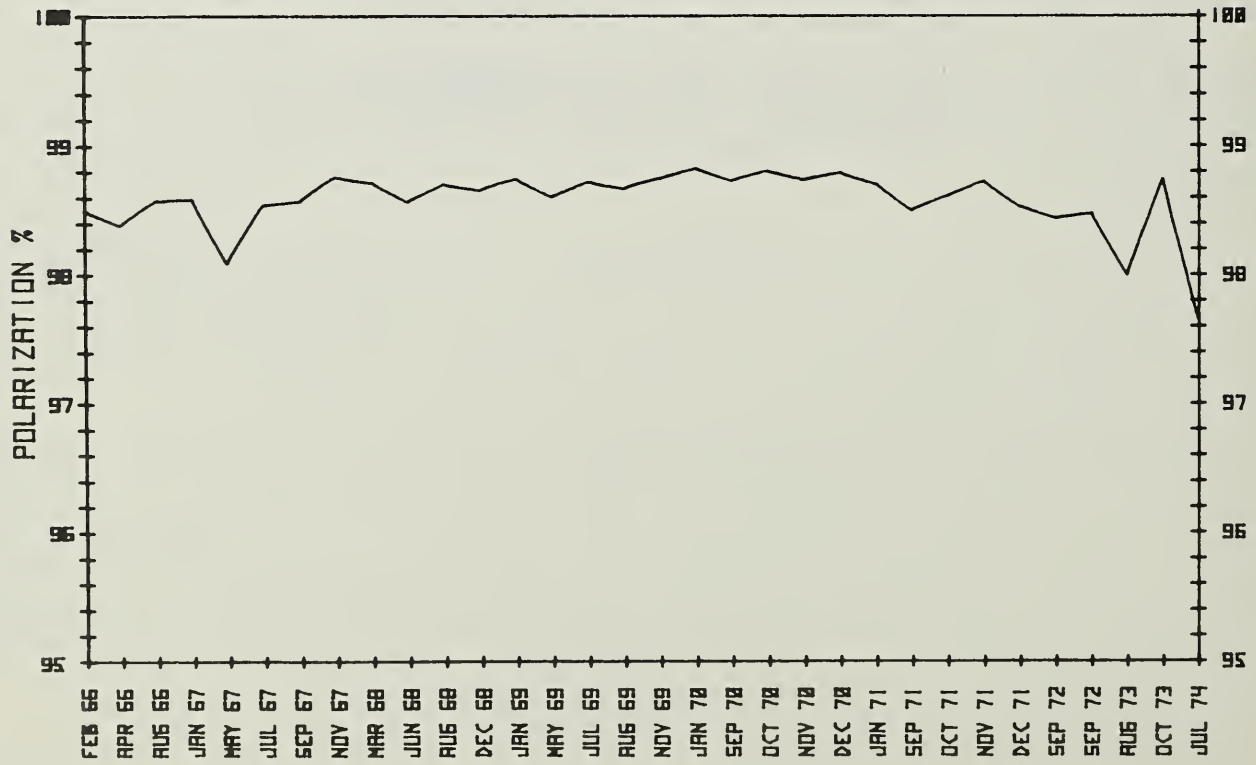
By the Staff of The British Columbia Sugar Refining Co. Ltd.[1]

(Presented by F. G. Carpenter)

Generally, the B. C. Sugar Refining Co. has observed little change in the quality of raw sugar from specific areas. Starch used to cause filtration problems but nowadays enzymatic treatments at the raw sugar mills have effectively eliminated that particular problem. B. C. Sugars has a higher output of soft sugars than the average refinery, which influences the raw sugar requirements somewhat. The raw sugars that were processed by the Vancouver refinery during the past ten years can conveniently be split into three groups according to origin. During the first half of the ten-year period, preferential duty sugars were mostly obtained from Fiji so that these raws really make up the first group. During the second half of the ten-year period the Fiji raws were predominantly replaced with raws from Queensland so that these raws represent the second group. The third and final group of raws, which were processed sporadically during the whole ten-year period, were the full duty non-preferential raws. These raws originated from many different countries so that for the purposes of this survey they have been collectively identified as non-preferential raws. Ten-year trend graphs for each of seven routine analytical components, polarization, moisture, invert, ash, insoluble impurities, organic non-sugars and crystal colors, have been prepared for the raw sugar groups. The graphs are self explanatory and require no further comments.

RAW SUGAR-FIJI

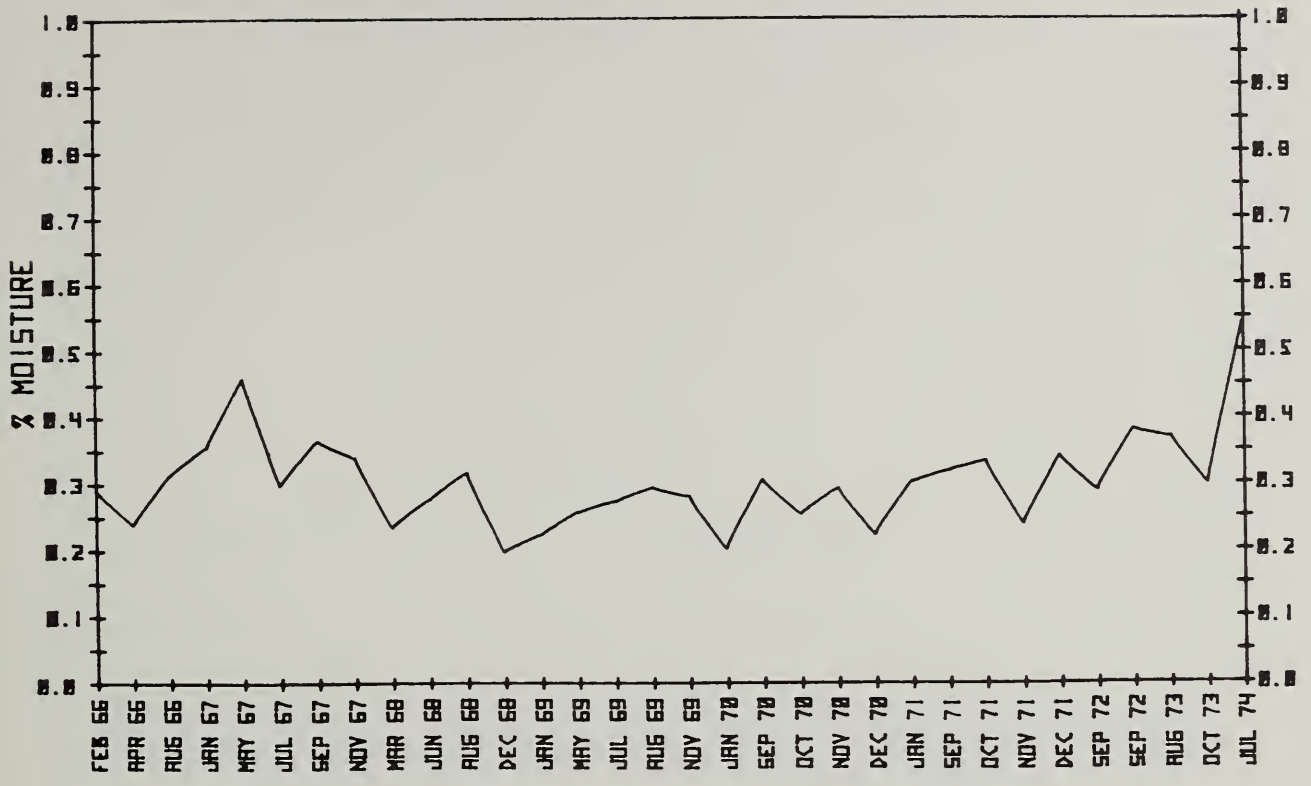
10 YEAR ANALYSIS TREND



[1]British Columbia Sugar Refining Co. Ltd., P.O. Box 2150, Vancouver B. C., CANADA V63 3V2.

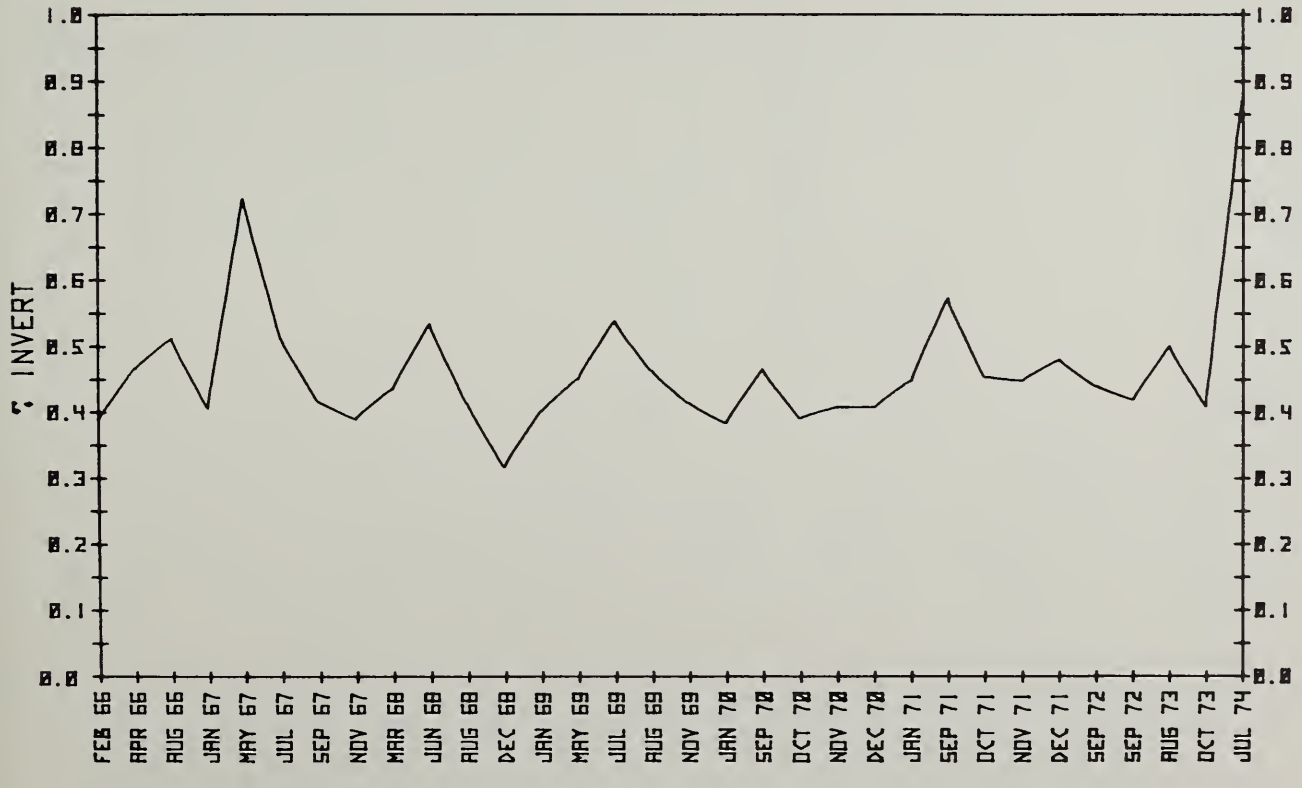
RAW SUGAR-FIJI

10 YEAR ANALYSIS TREND



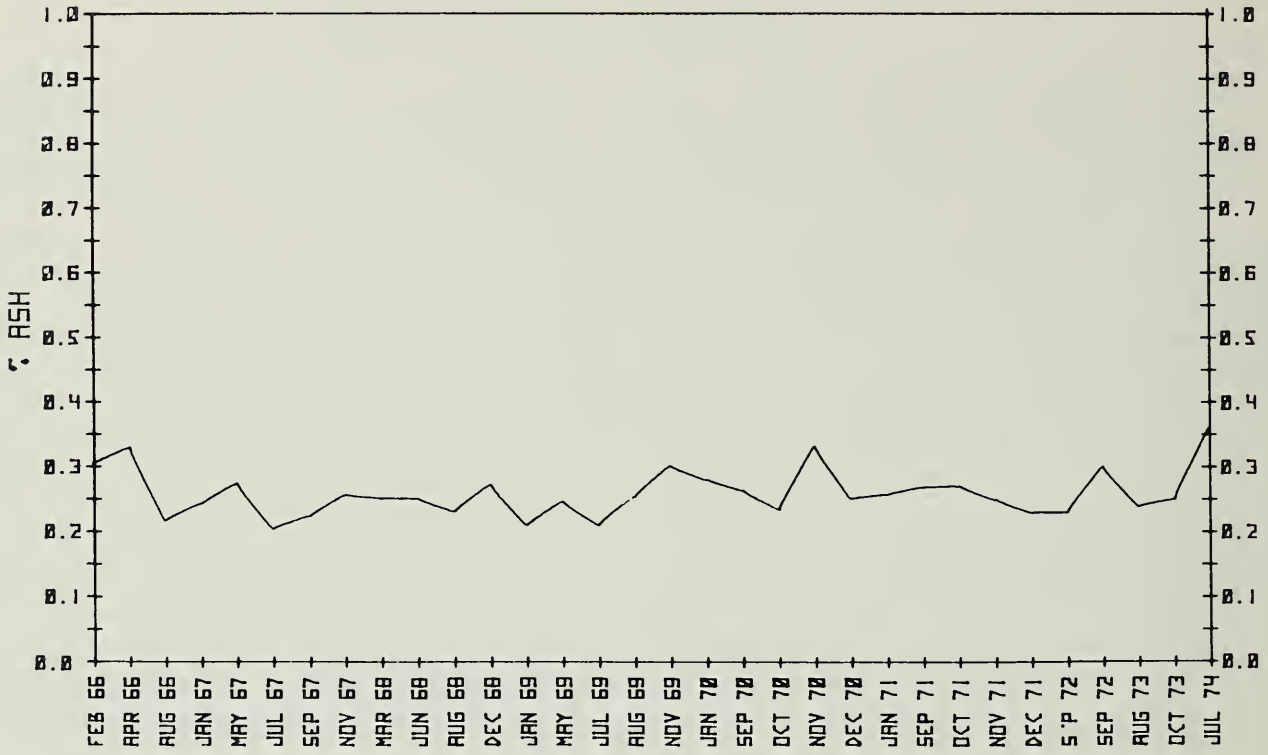
RAW SUGAR-FIJI

10 YEAR ANALYSIS TREND



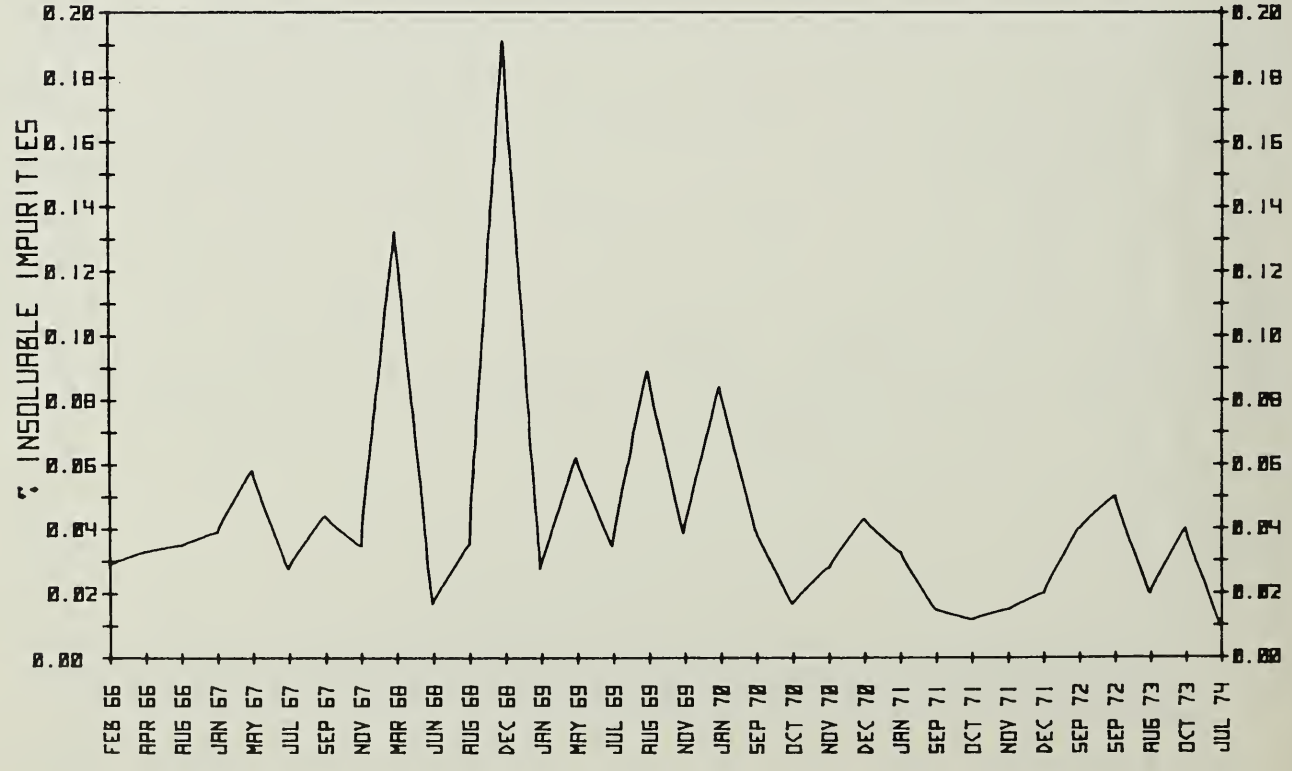
RAW SUGAR-FIJI

10 YEAR ANALYSIS TREND



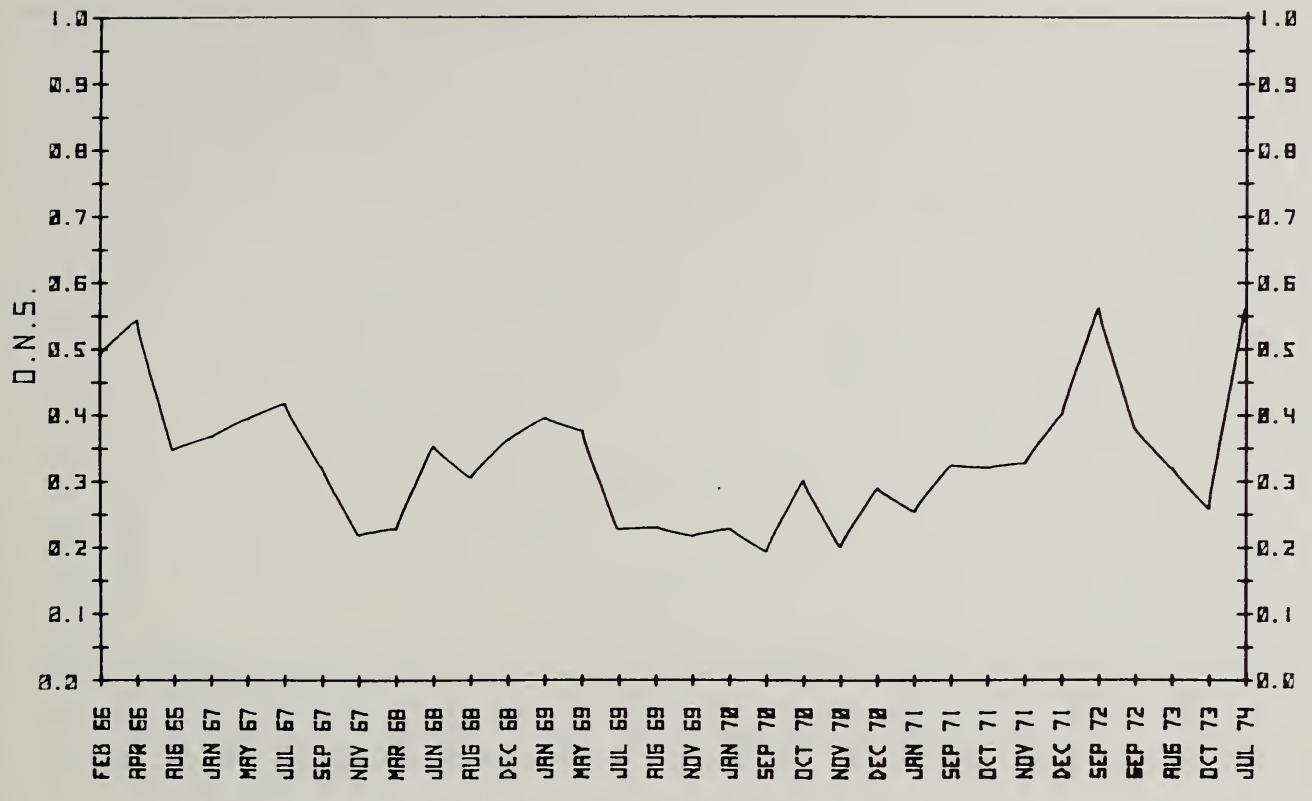
RAW SUGAR-FIJI

10 YEAR ANALYSIS TREND



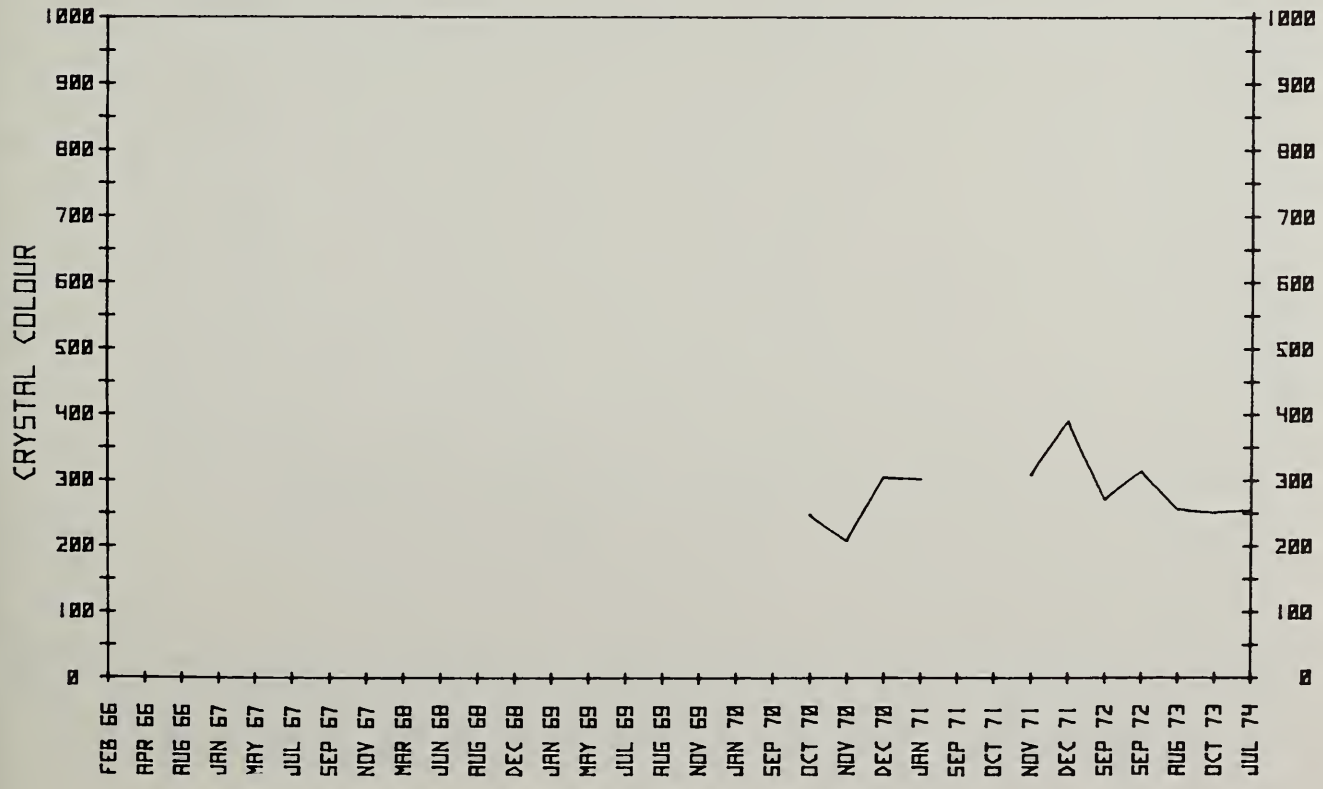
RAW SUGAR-FIJI

10 YEAR ANALYSIS TREND



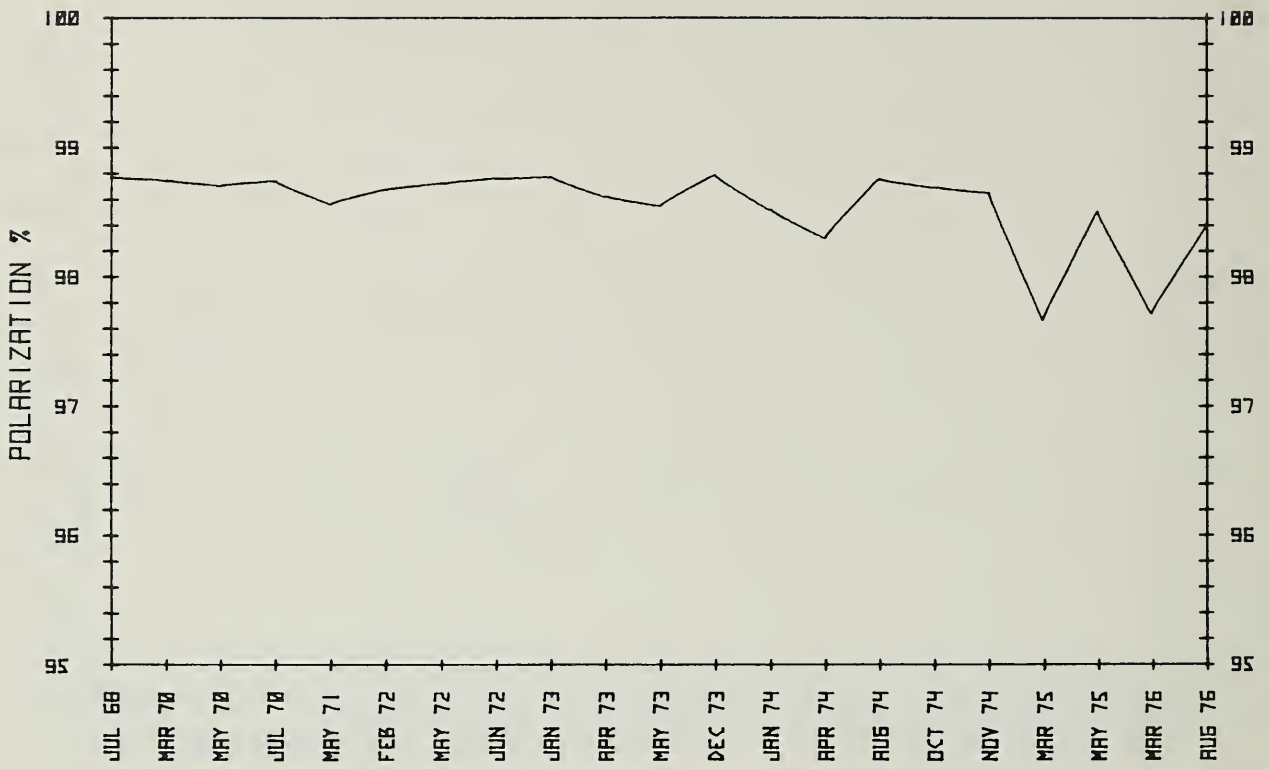
RAW SUGAR-FIJI

10 YEAR ANALYSIS TREND



RAW SUGAR-QUEENSLAND

10 YEAR ANALYSIS TREND



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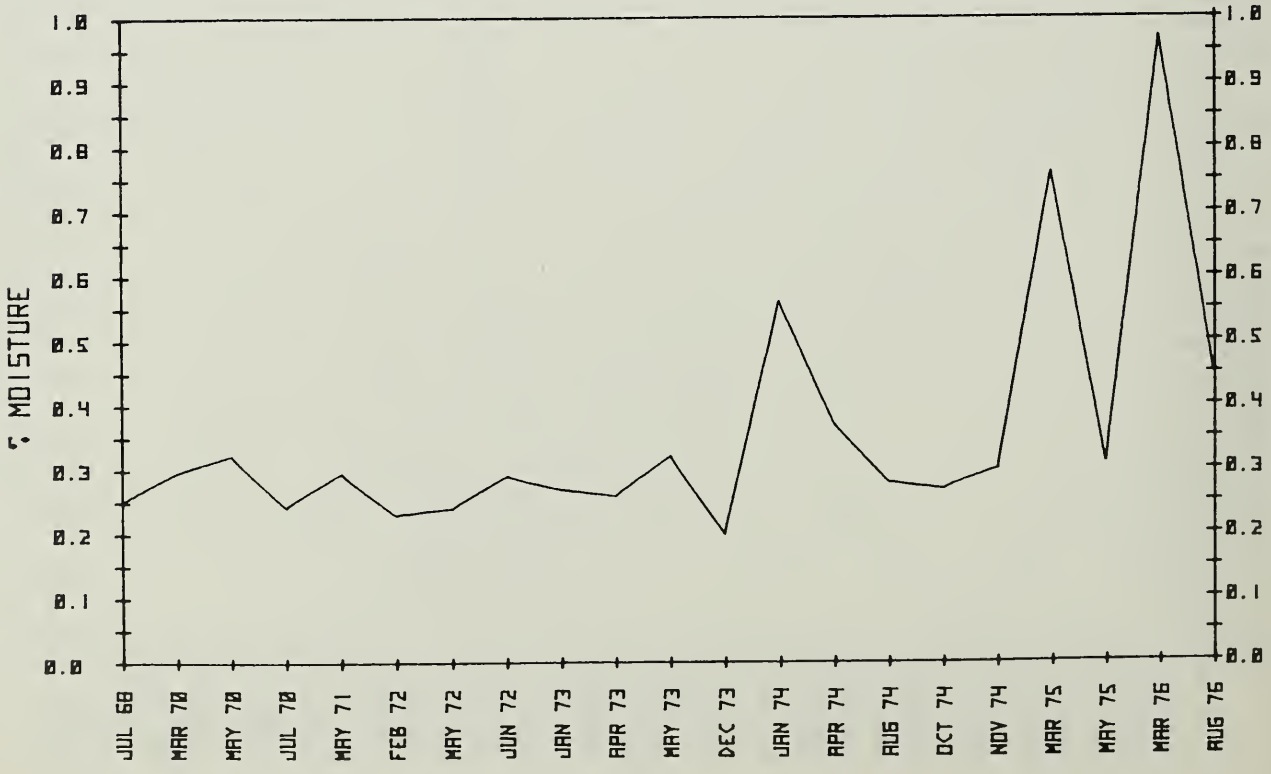
10 YEAR ANALYSIS TREND



RAW SUGAR-QUEENSLAND

10 YEAR ANALYSIS TREND

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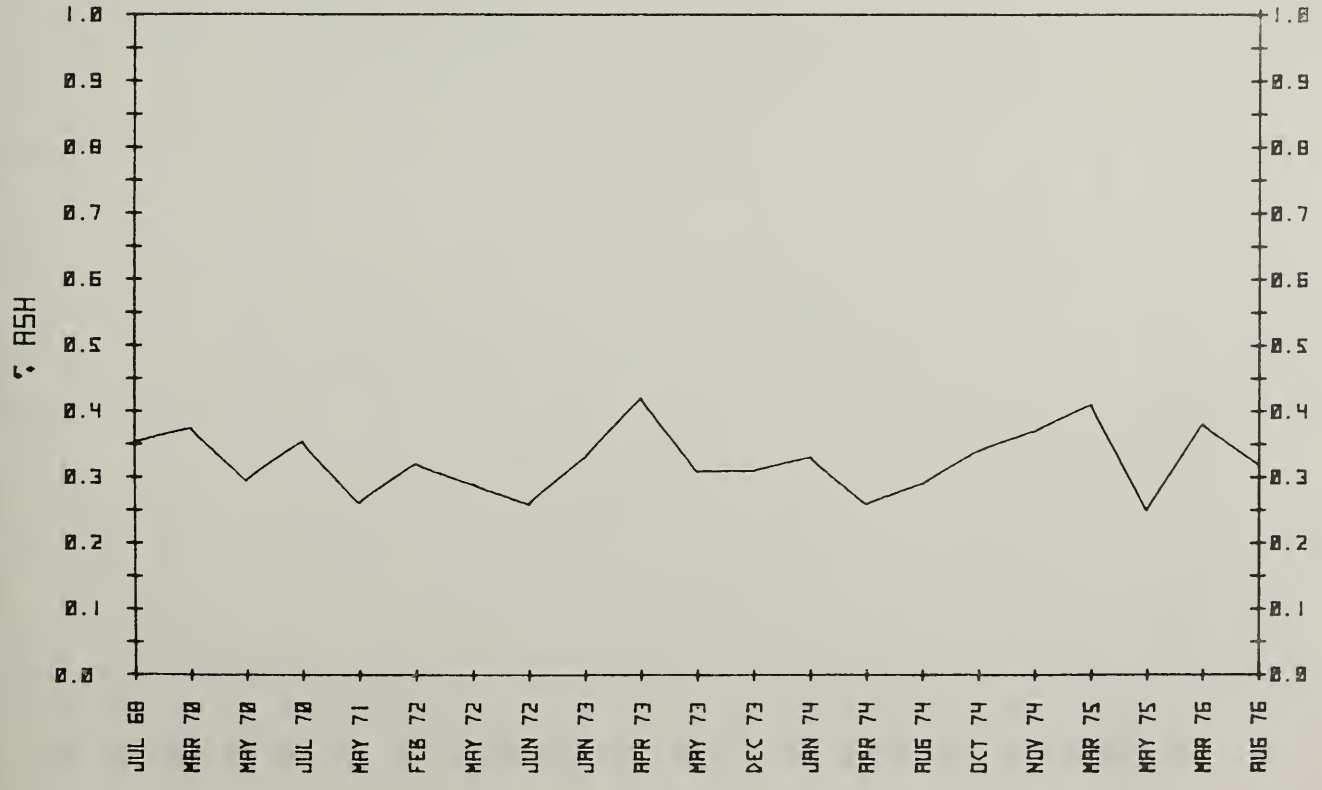

RAW SUGAR-QUEENSLAND

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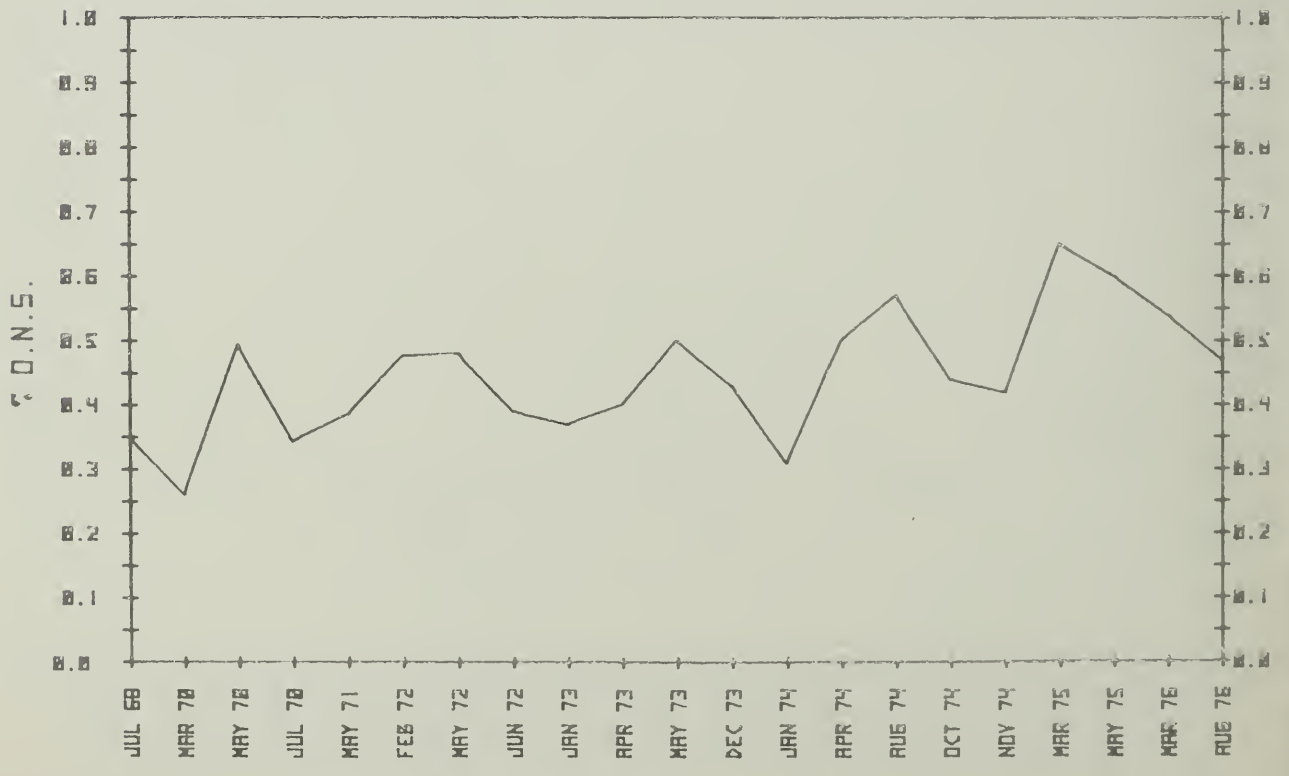
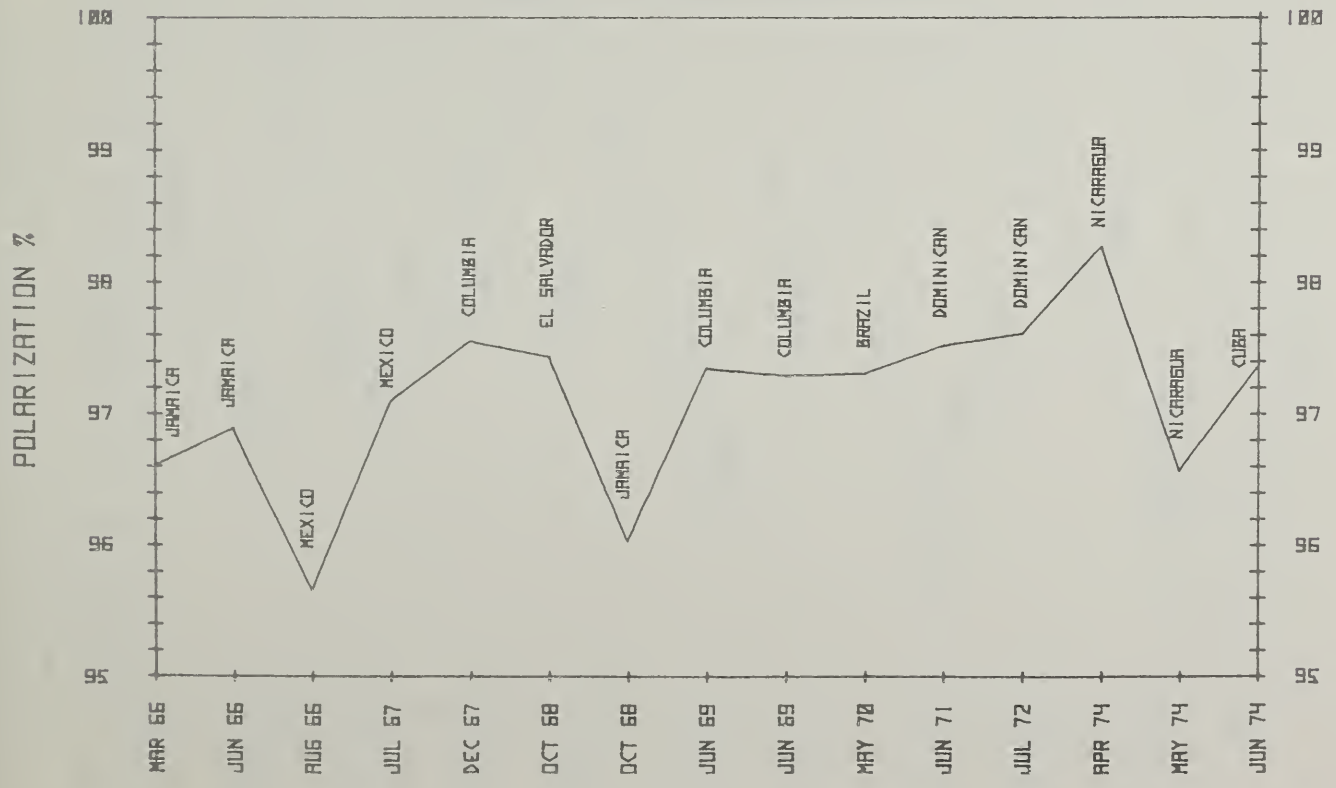


RAW SUGAR-QUEENSLAND

10 YEAR ANALYSIS TREND

RAW SUGAR-NON PREFERENTIAL

10 YEAR ANALYSIS TREND



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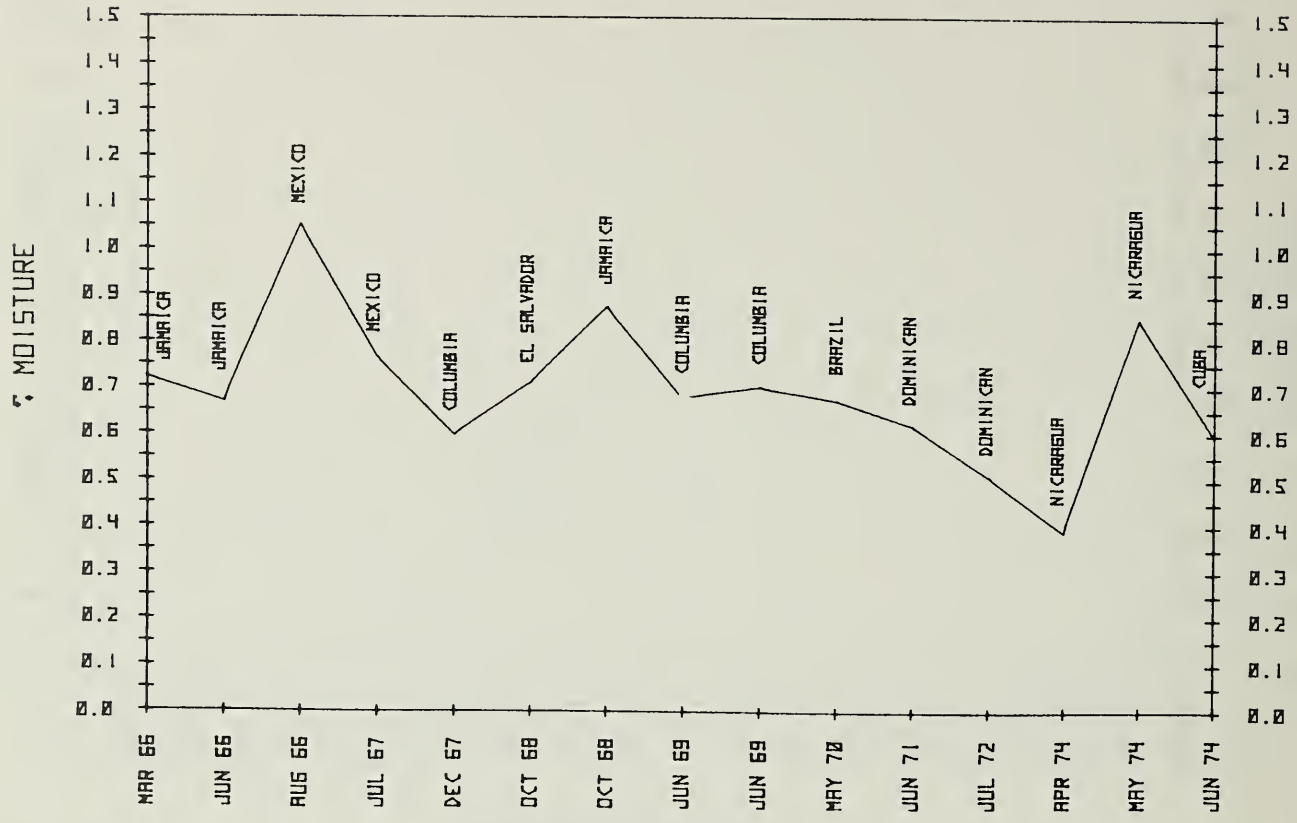

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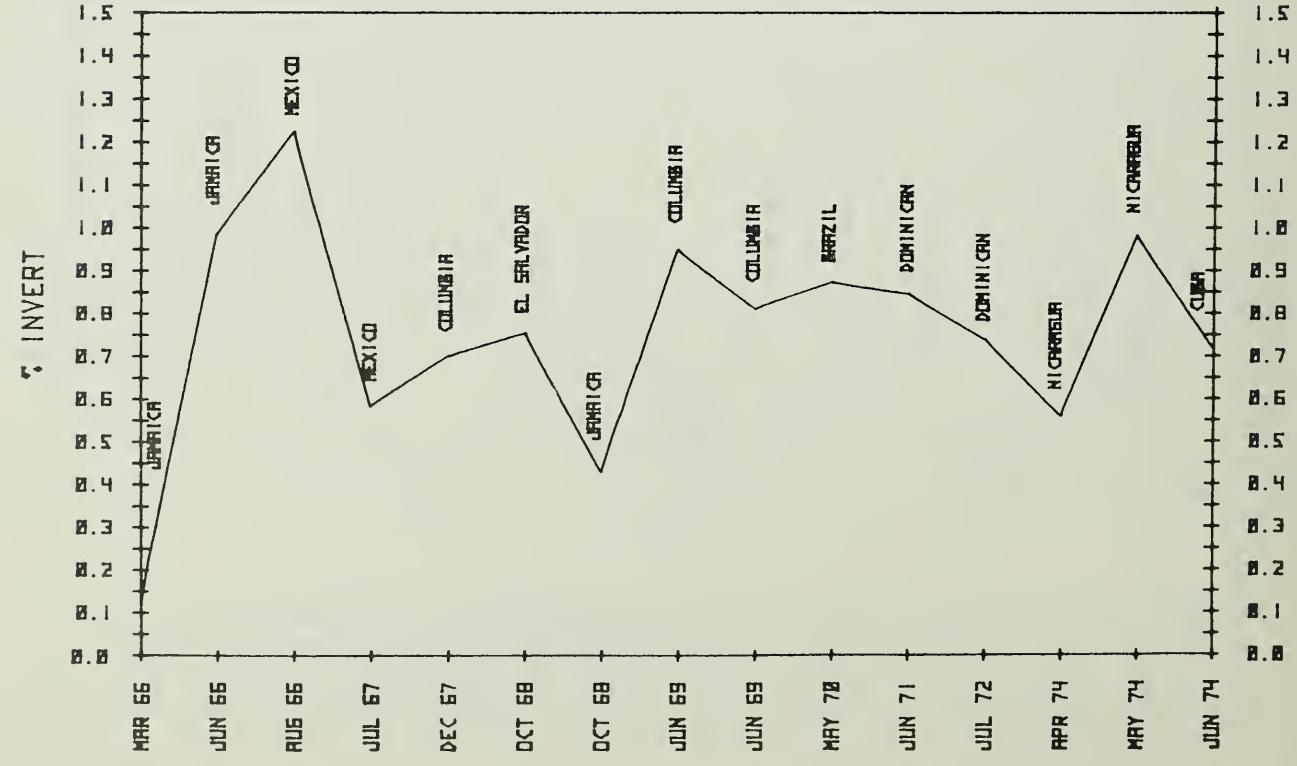

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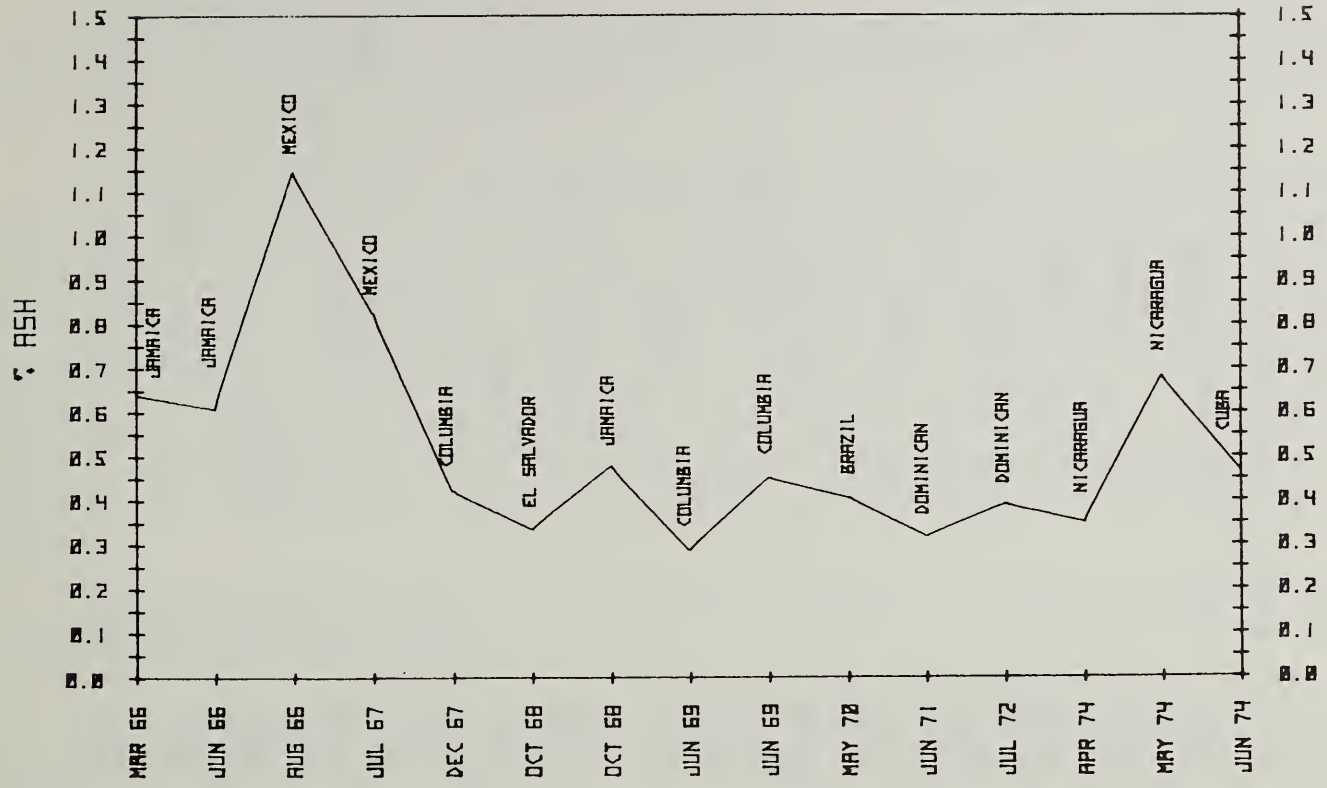

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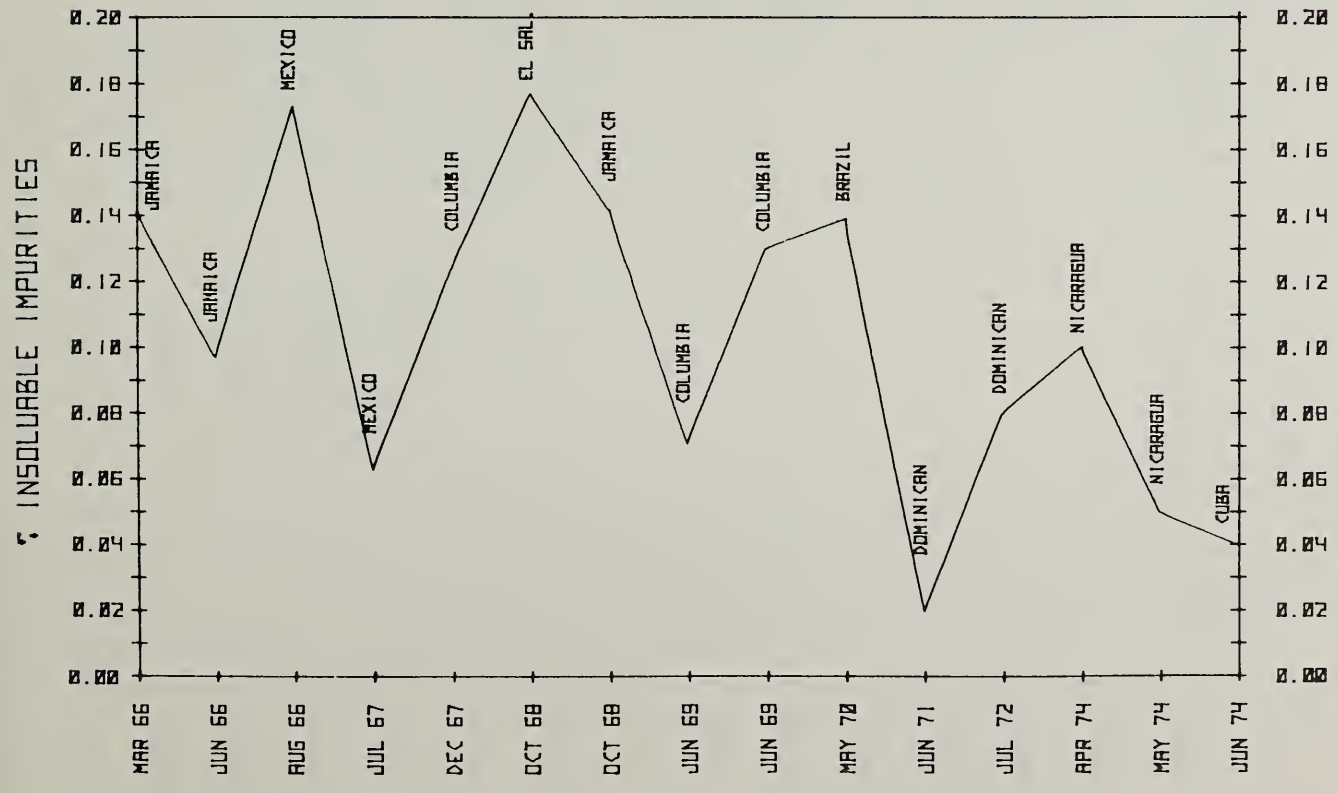
10 YEAR ANALYSIS TREND



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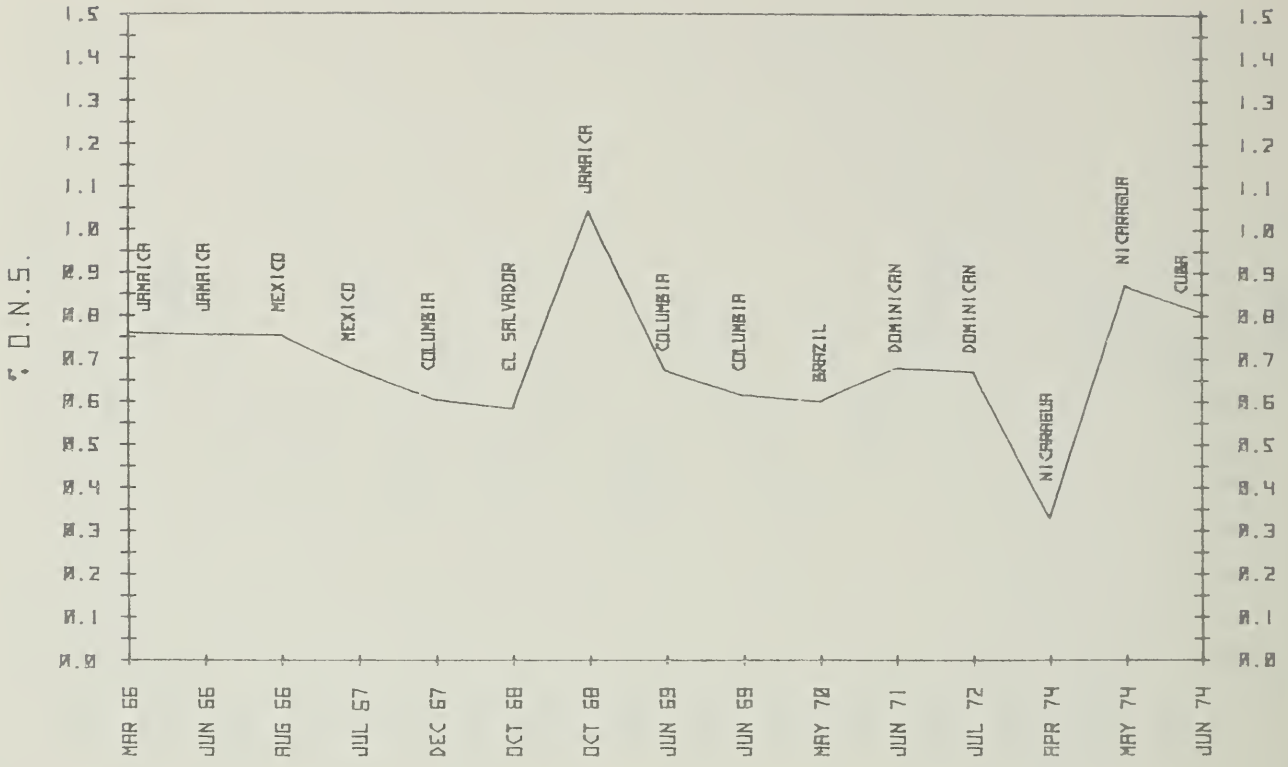

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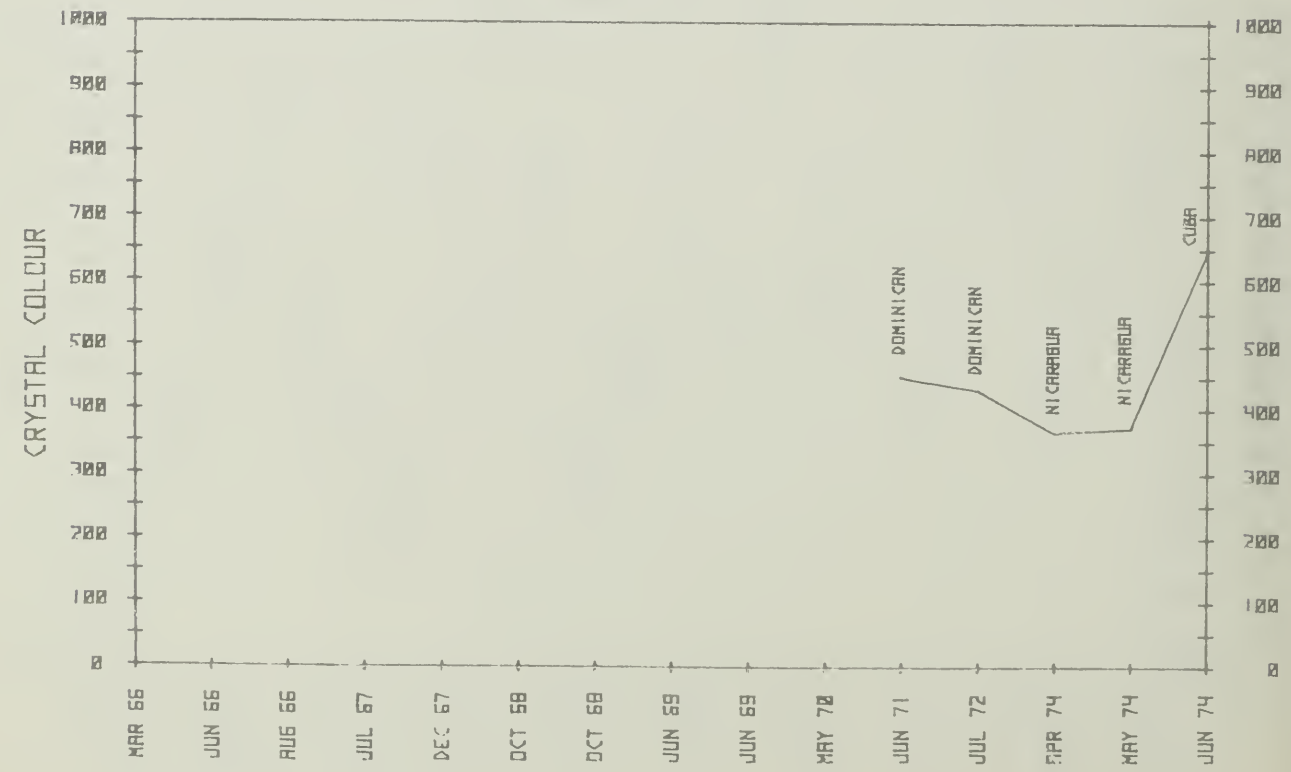

H. G. Gerstner: We deduce from Mr. Lopez-Ona's remarks that he is not wildly enthusiastic about the penalty and premium feature of the quality standards. At least one other member of this panel privately said, using technical terms, that they are not worth a damn.

R. Moroz (Sucrest): You stated that one of the reasons why a raw sugar producer doesn't produce on a consistent basis to stay in the premium area is that most are more concerned with raw sugar prices than quality standards. I think that if you applied any of these parameters against world sugar prices you would see a correlation with this up and down cycle. In 1974 and '75 the prices were sky high, and the quality was quite poor. So, the raw sugar producers are certainly more involved with prices than with quality. I think the penalties are not great enough to give the incentive. Also, there are government policies in many of these countries on whether to produce or not produce particularly for export. In 1976 and 1977 you will see very little raw sugar from Brazil and Mexico.

J. V. Lopez-Ona: When I spoke to them in 1976 they were not talking about 1974, 1975, they were considering 1976.

J. F. Dowling (Refined Syrups): Ten years ago when Jim Culp set these standards up, it was a massive task. Five years after that we did try to change the ash to the washed raw sugar ash, which means a lot more to us and to a lot of refineries. We have had that for 10 years and it has undoubtedly served its purpose. The initial thrust was great; getting the standards off the ground was tremendous and now it is up to us to improve them.

There is no question but that if someone comes to me and says, I am getting a cargo from say, Mars, I do not look at the raw sugar quality standards that we talked about today. The first thing I look at is the ash on the washed raw sugar because we are an ion-exchange refinery. The second thing is the floc test. What I look at, and what probably most of the people sitting in this room look at was not discussed today. We should discuss what we think most important even if it is not on the list of present sugar quality tests. I don't get too excited and argue with the raw sugar buyer if he has a dollar off because the grain size is slightly different, but we would argue with him if we knew we were going to have floc coming out of his sugar. Maybe we ought to look at what we can tell the sugar producer that we consider even more critical, than some of the things we have listed.

The other thing that we should consider are the particular questions our customers ask: is there asbestos in the product? any heavy metals? Do we ask this of the raw sugar producer? Do we ask what was in that cargo ship when he loaded it? Was it loaded previously with lead oxide? These are the type of questions that we ought to spend a little more time on. Conversation with the raw sugar people is probably more important.

W. R. Tuson (Colonial Sugars): The comments so far have brought out many interesting points for all of us. I have some general comments. I think the institution of quality standards of raw sugar was a very fine idea. During the time it has been before us, we have gained a lot of background data information with respect to raws of various origins, which we ought to put to good use. However, actually I don't think that it has had much effect, speaking from the viewpoint of Colonial Sugars. The origins that we considered producers of good sugar prior to installation of quality standards are still good. Those which were considered poor are still poor; those that were intermediate are still intermediate. This brings to a head the point that was previously made that premiums and penalties have had very little influence. I can only think of one particular origin that had a definite change in their sugar marketing policy, and their quality is worse. I'd like to go back to the comment I made at the first symposium on Raw Sugar Refining Quality[1]. That is the fact that in dealing with standards, we have looked at these standards as individual items and we seem not to have put any weight on the interrelationship, on one quality criterion vs. another. I'm thinking particularly of color-ash relationship, which if you remember, I commented on at the last symposium . Color and ash have a interrelationship in the refining process as was pointed by Carpenter & Deitz in the 50's with their Excess Polyvalent Anion Theory[2]. I think we cannot look at these two things as individual characteristics. I think we have to find some basis of joining them together, because I have seen raws which could bring a color quality premium, whose ash values were not in a penalty range but were on the high side. The value of that color premium that we paid was zero, because of the effect of ash, in conjunction with whatever color was in the raw, which made the apparently low color difficult to remove.

J. A. Harrison (Supreme): In the last symposium in 1968[1], I stated that we at Supreme, because a goodly portion of our sugars comes from Louisiana, were in a position to do something about setting standards. We initiated a penalty and premium program two years before Contract No. 10. Ours worked, and I think because we had feedback. The proximity of the mills to us was such that we could tell them what problems were arising, and what was successful. We are happy to report that we discontinued the penalty and premium program with our Louisiana suppliers - with the exception of one standard - temperature - we set it at 103°F. We had the same problem with color formation on storage that they have at Savannah before we did that.

So in general - if there were feedback, I think that the whole raw sugar quality standards would work, but I agree with others that the incentives are not quite right.

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[1]Symposium, 1968. Raw sugar quality standards. Proc. Cane Sugar Refin. Res. 1968: 138-159.

[2]Carpenter, F. G., Larry, D., and Deitz, V. R. 1961. Ionic interactions with sugar colorant during char filtration, Tech. Sess. Bone Char 7:259-292.

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